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CATALYSIS I

TOWARD A GREEN SYNTHETIC INORGANIC CHEMISTRY

Yi Lu

Assistant Professor, Department of Chemistry, University of Illinois at Urbana-Champaign, Urbana, IL 61801

ABSTRACT

Inorganic and organometallic compounds are widely used as powerful catalysts in industrial processes. Yet most of them are not considered environmentally benign due to the toxicity of organic solvents used and non-biodegradability of the compounds. Similar problems occur in the field of biomimetic synthesis; while many synthetic models can mimic either the structural or functional features of native enzymes, few models can duplicate the environmentally friendly virtues of enzymes since non-water-soluble and non-biodegradable organic molecules are used as building blocks.

We are interested in overcoming these problems by creating similar metal-binding centers inside a small, stable, and well-characterized protein. Our approach is a natural extension of the traditional inorganic synthesis, except that the ligands are proteins rather than small organic molecules. Our syntheses are carried out under physiological conditions, using largely non-toxic reagents in aqueous solution. The products are also biodegradable.

This approach has been applied successfully in making a biomimetic, environmentally friendly model of manganese peroxidase, a heme peroxidase that is active in biodegradation of lignin (the second most abundant bio-feedstock) and bioremediation of aromatic pollutants (such as polychlorinated biphenyls). More importantly, the combination of the power of metal ions' ability to control reactivity with the ability of proteins to guide regio-, stereo-, and enantio-selectivity allows us to make new catalysts that are better than either traditional inorganic catalysts or enzymes. Recent progress in this direction, and the advantage and disadvantage of our approach over traditional chemical and biochemical approaches will be discussed.

INTRODUCTION

Synthetic inorganic chemistry has played an instrumental role in the advancement of science and engineering by providing new, efficient, and durable catalysts and materials. However, inorganic syntheses are normally carried out in toxic organic solvents using non-biodegradable organic molecules as building blocks. To overcome these problems, several novel methods have been used^{1, 2}, including designing efficient reactions with atom economy and in aqueous solution. In order to make synthetic inorganic chemistry more environmentally benign, new and creative approaches are needed. We can search nature for answers since metalloenzymes and biomaterials are environmentally benign *and* are superior to many of the man-made catalysts and materials. While many biomimetic models have been synthesized that can mimic the native enzymes either structurally or functionally, few models are made as environmentally safe as the enzymes. It has been difficult to mimic some of the features in biological systems, such as site-specific modulation of the secondary coordination sphere, and the regio-, stereo-, and enantio-selectivity of the system. The key difference between traditional inorganic chemical synthesis (including biomimetic synthesis) and biosynthesis is that the former uses non-water-soluble and non-biodegradable organic molecules whereas biosynthesis utilizes proteins.

We have been using stable, easy-to-produce, and well-characterized proteins as a scaffold for designing and engineering artificial metalloenzymes that either have similar structural and functional properties to much more complex native enzymes, or possess new structure or reactivity that is unprecedented in nature³⁻⁹. The key to the success of our approach is to take advantage of the recent developments in molecular biology techniques such as the polymerase chain reaction and improvements in protein expression and purification methods. Typically, our lab completes the design, construction, and purification of an enzyme in 10-14 days to yield 20-250 mg/L of highly purified enzyme⁴⁻⁹. These numbers are quite comparable to synthetic model synthesis. Furthermore, all proteins in our studies have been well-characterized by spectroscopy and are amenable to NMR and crystallization studies. The combination of the high catalytic reactivity of a metal ion with the rigid and chiral environment of a protein allows us to design and engineer a new generation of catalysts with reactivity and selectivity unprecedented in the field. In addition, through a systematic variation of the protein environment around both the metal-binding and substrate-binding sites, we can tailor the reactivity and regio-, stereo-, and enantio-selectivity with high precision. Since proteins are hydrophilic outside and hydrophobic inside, many transition metal catalysts that are not normally soluble in water can be made to be functional in aqueous or aqueous/organic environment. These features, together with the biodegradability of the products, make this an environmentally benign approach.

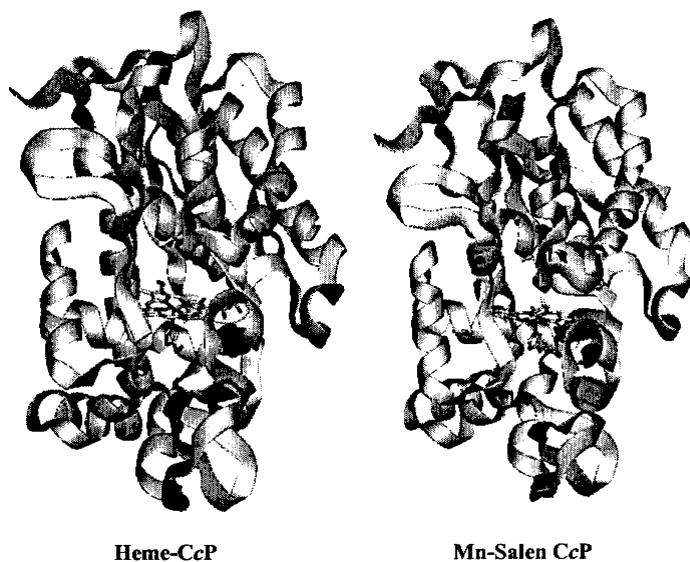
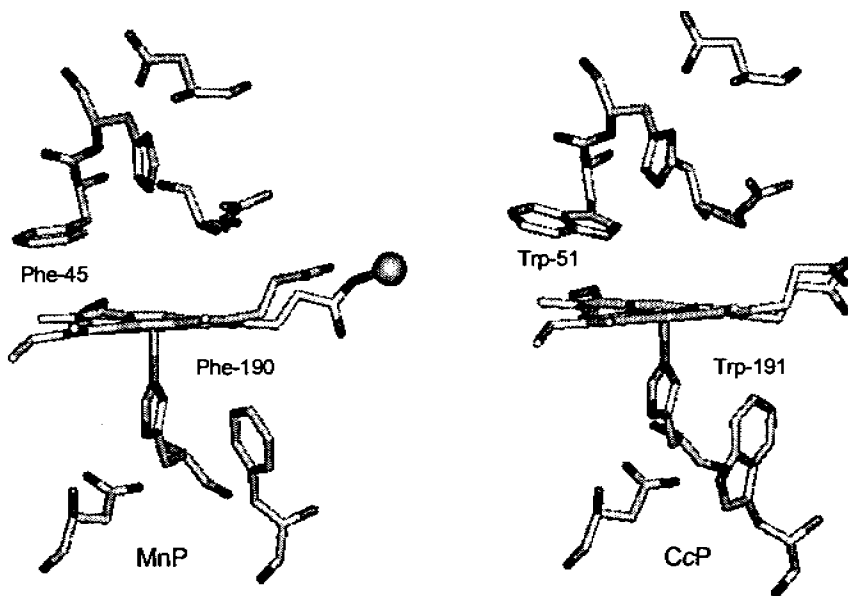
RESULTS AND DISCUSSION

Design and Engineering a Heme Peroxidase for Biomimetic and Environmentally Safe Biodegradation of Lignin and Bioremediation of Aromatic Pollutants.

Manganese peroxidase (MnP) is a key enzyme in lignin degradation system of the basidiomycetous fungus *Phanerochaete chrysosporium*¹⁰. It is involved in biodegradation of lignin and bioremediation of aromatic pollutants, such as dibenzodioxins and polychlorinated biphenyls. Controlled lignin degradation could provide renewable energy sources since lignocellulosic materials, the most abundant renewable materials on earth, could be converted to chemicals and fuels. The ability to degrade aromatic pollutants would allow us to dispose of this group of toxic wastes. We have successfully designed and synthesized a biomimetic model of MnP using cytochrome *c* peroxidase (CcP), a heme peroxidase with less than 25% sequence homology and no MnP activity⁷⁻⁹. We accomplished this task by engineering a Mn(II)-binding site into CcP and by changing the environment around the heme center. Spectroscopic studies demonstrate a striking similarity between our model and the native MnP^{7, 8}. Dramatic increase in the MnP activity is also observed in the designed proteins, one of which possesses MnP specific activity that is within one order of magnitude of the native MnP^{7, 9}.

Engineering Artificial Metalloenzymes for Asymmetric Catalysis.

Mn(III)(Salen), an achiral version of the well-known Jacobsen's catalysts for asymmetric epoxidation, has been incorporated into CcP, a protein that can be engineered and purified with high yield. The substrate-binding sites are being engineered by rational design using computer modeling and site-directed mutagenesis or by random mutagenesis and *in vitro* selection. The combination of both approaches will allow us to employ negative controls (such as steric blocking to disfavor the formation of the unwanted enantiomer) and positive controls (such as specific interactions with the substrates using H-bonding, π - π stacking, and electrostatic interactions) on the regio-, stereo-, and enantio-selectivity. The turnover number of this class of catalysts is increased dramatically by using proteins as ligands to protect the catalysts. Finally, we are exploring ways to stabilize the enzymes during organic transformation.



Engineering Novel Bioorganometallic Enzymes.

Organometallic complexes are very powerful and highly versatile catalysts for many chemical reactions such as C-H bond activation and hydrogenation. Many catalysts currently used in industrial processes are based on organometallic complexes. The combination of an organometallic center with a protein scaffold can result in a potentially superior biocatalyst that is both highly reactive and selective. Cobalamins in Vitamin B₁₂ are well-known organometallic centers found in biological systems. Recently, another organometallic center was discovered in carbon monoxide dehydrogenase. While it remains to be seen if more organometallic centers will be discovered in biological systems, building an organo-metallic enzyme represents an interesting challenge that, if successful, will have a fundamental impact on catalysis in both chemistry and enzymology. We have prepared a

ferrocene-cytochrome *c* peroxidase (Fc-CcP) derivative in which ferrocene is covalently linked to heme-free CcP. Upon oxidation to ferrocenium, the Fc-CcP can replace the native heme-containing CcP by oxidizing cytochrome *c*. Since redox potentials of ferrocenes can be tuned as widely as 1200 mV by different substitutions on the Cp ring, the resulting Fc-CcPs can be used as artificial, water-soluble electron transfer proteins. Other organometallic centers are being incorporated into proteins so that a new generation of asymmetric-organometallic catalysts can be made.

SUMMARY

Our results demonstrate that introducing proteins into traditional synthetic inorganic chemistry not only makes inorganic chemistry greener, but also better. Encapsulation of inorganic and organometallic compounds into proteins provides a way to transform inorganic catalysts into asymmetric and more environmentally benign catalysts. It is our belief that the full potential of our approach is yet to be realized. We hope the concept developed in these studies may serve as a basis for further investigation.

ACKNOWLEDGEMENT

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ENVIRONMENTALLY BENIGN PROCESSES FOR THE PREPARATION OF NOVEL POLYMERIC MATERIALS BY CONTROLLED/"LIVING" RADICAL POLYMERIZATION

Scott G. Gaynor

Assistant Director - Center for Macromolecular Engineering

Krzysztof Matyjaszewski

J.C. Warner Professor of Natural Sciences, Department of Chemistry
Carnegie Mellon University, Pittsburgh, PA 15213

The development of novel polymeric materials has been the goal of every synthetic polymer chemist since Staudinger and Carrothers first established the premise that polymers were "macromolecules" comprised of a large number of repeating smaller molecules, monomers. The early days of synthetic polymer science focused on the development of new monomers which were capable of being polymerized to form useful polymeric materials. After a sufficient number of polymeric materials were identified, work then turned towards understanding the properties of these materials and later to establish structure property relationships between monomer and the final, bulk, polymeric material.

The search for, and development of, new monomers was at first a major emphasis in polymer chemistry, but soon faded in importance. As the properties of simple monomers were understood, as well as a better understanding of how polymers behaved as materials, the need for new monomers waned. However, new monomers are still being developed with the expectation that vastly different properties from existing monomers will be attained.

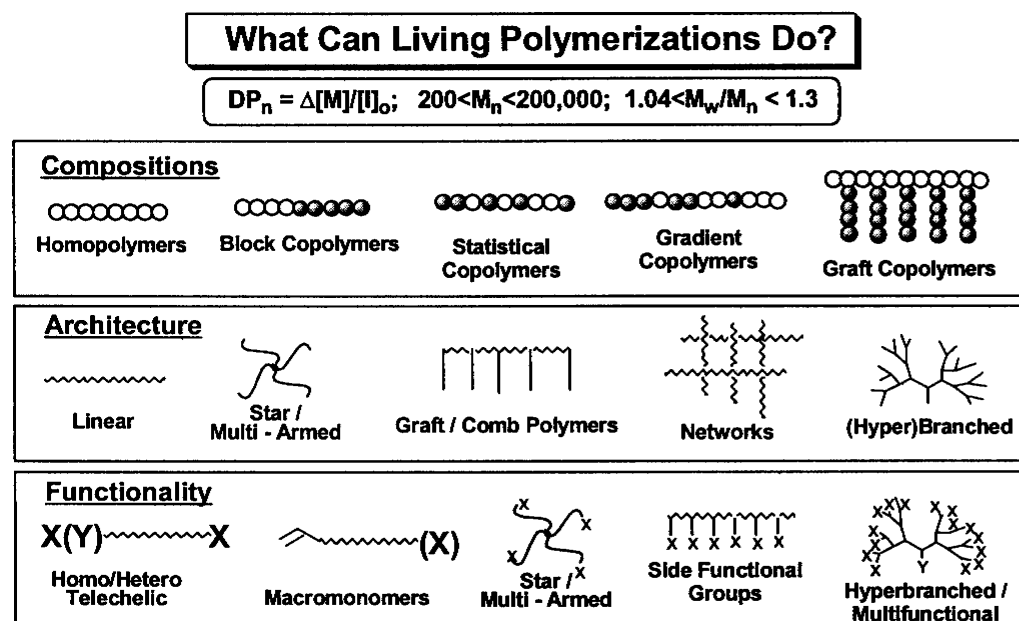
The decrease in demand for new monomers was partially the result of better understanding of the physical properties of polymers and how to exploit those properties. As new polymer engineering principles and processes were developed, enhanced properties could be extracted from the simplest polymers. Additionally, advances in simple chemical modifications of base polymers and in varying formulations of these base polymers with small molecule additives, polymers with very novel and specific properties were attained. Although all of these advances have been significant and have allowed for polymers to be ubiquitous in our everyday lives, these methods generally result in incremental advances; significant or quantum leaps in polymer properties are generally not observed.

To make polymeric materials with significantly different, if not better, properties, one must either return to the expensive undertaking of developing new monomers, or find new methods of polymerizing those monomers so that novel properties can be achieved. Towards the latter, polymer chemists have striven to develop living polymerizations.

These polymerizations are not living in the biological sense of the word, but refer to systems where all polymer chains grow at relatively the same rate, and that none are irreversibly terminated, i.e., can no longer add monomer and grow. Living polymerizations were first reported in 1956 with the anionic polymerization of styrene.¹ Subsequently, living polymerizations were extended to cationic, ring opening metathesis polymerization (ROMP) and Ziegler-Natta polymerization systems. Living polymerizations offer the ability to control not only

molecular weight (M_n) and molecular weight distributions (M_w/M_n), but also the composition, architecture and functionality of the polymers, Figure 1. When compared to other polymerization systems that afford only homo- or statistical linear polymers, one can see that many new and different polymers can be prepared and their physical/mechanical properties exploited.

Figure 1. Microstructures possible by living polymerizations.



Until only very recently have systems been developed which can allow for the preparation of a controlled/"living" radical polymerization (termination is never truly avoided, but is suppressed) that offers the promise of overcoming all of the drawbacks of other living polymerizations.² Our group has focused on using a metal

catalyzed system called atom transfer radical polymerization, ATRP.^{3,4} ATRP has shown much promise as a controlled/"living" polymerization system because of its ability to polymerize a wide range of monomers (styrenes, acrylates, methacrylates, acrylonitrile, etc.) using inexpensive alkyl halide initiators and transition metal catalysts. ATRP has been used to prepare all of the various compositions, architectures, and functional polymers described in Figure 1.

While ATRP offers the ability to prepare well defined polymers and novel polymeric materials, it also allows for the production of environmentally friendly products by environmentally friendly processes. Conventionally, radical polymerizations are conducted in solutions consisting of monomer and organic solvent; they may also be conducted in bulk, but only to relatively low conversions. The reason for these limitations is that the reactions may run out of control (auto-accelerate) and cause undesired reaction conditions, or worse, explosions. To some degree, these problems have been overcome by emulsion polymerization where the reaction is conducted in water, generating latexes which can be used directly unless the polymer must be isolated, which requires additional process steps. ATRP does not have these problems of auto-acceleration, and can be conducted in bulk to nearly complete conversion of the monomer. Thus, it is possible for the polymer to be used directly without the addition of VOCs as diluents. As an added benefit, ATRP can also be conducted in emulsions, allowing for the production of well-defined polymers in water.^{5,6} ATRP has also been successfully conducted in liquid or supercritical carbon dioxide.⁷

The products that are produced by ATRP can also be used to alleviate other environmental/health problems. For example, it would be desirable if coatings could be applied without the use of VOCs as solvents. Unfortunately, current systems employ high molecular weight resins, which must be diluted in order to obtain good flow properties during application to surfaces. To overcome this, ATRP can produce low molecular weight, functionalized polymers that easily flow without added solvent and can then be reacted together during application to give high gloss, chip-resistant and durable coatings. Thus, VOCs can be reduced or eliminated from coatings applications by use of ATRP prepared polymers.

Another example is in the use of plasticized poly(vinyl chloride) (PVC). PVC normally is very brittle and additives (plasticizers) must be added; commercial PVC contains 20-45% plasticizer. As this plasticizer can leach out over time resulting in the PVC becoming brittle, it must be periodically replaced using products such as ArmorAll®. Of even greater concern is the leaching of phthalate plasticizers from children's toys and products that come into contact with body fluids, e.g., IV bags. There have been recent reports that phthalates may cause cancer; obviously the use of a flexible PVC that does not use phthalates would be beneficial to society. To solve these problems, ATRP has been used to prepare a PVC graft copolymer that has soft poly(butyl acrylate) (PBA) chains covalently attached to the PVC chains.⁸ The PBA acts as a plasticizer, but since it is bound to the PVC chain it can not leach out. Such graft copolymers can be used directly or be used as compatibilizers for simple blending of PVC and PBA; PBA is a high molecular weight polymer, is completely nontoxic and FDA approved.

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TOWARD BENIGN SYNTHESIS VIA CATALYTIC OXIDATIONS USING DIOXYGEN

Prof. Klaus H. Theopold, Sebastien Blanchard, Somying Leelasubcharoen,
Louise M. Liable Sands and Dr. Chandi Pariya

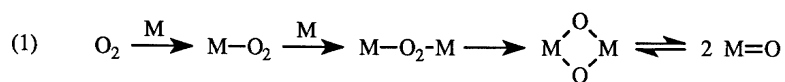
Department of Chemistry and Biochemistry, Center for Catalytic Science and Technology,
University of Delaware, Newark, DE 19716

ABSTRACT

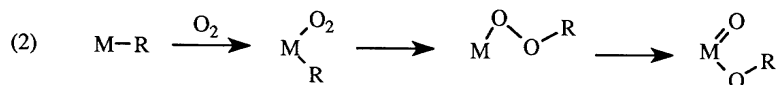
Oxidation reactions comprise a large class of industrially important chemical transformations. The use of dioxygen as a terminal oxidant is attractive from an environmental viewpoint, due to the lack of harmful byproducts. The principal problem with this approach - despite the considerable oxidizing power of O₂ - is the kinetic inertness of this molecule. Our research is concerned with activating dioxygen by binding to transition metals, and ultimately creating homogeneous catalysts for selective oxidations of organic molecules.

INTRODUCTION

In elucidating the fundamental mechanistic patterns of air oxidations of organic molecules, two complementary approaches can be taken. The first of these is to "activate" the dioxygen - i. e. transforming it into a reactive species - to the point where it will react in an intermolecular fashion with unactivated organic substrates. A possible sequence of transformations toward that end is depicted in equation 1. The crucial point appears to be that the O-O bond of O₂ must be broken; another is that the metal designated by M should be a late transition metal in a relatively high formal oxidation state.



An alternative approach is the activation of the organic substrate, for example via formation of an organometallic compound, which could subsequently react with O₂. Relatively little is known about the mechanisms of such reactions; equation 2 depicts a possible series of steps leading to oxygenative functionalization of a metal-carbon bond.

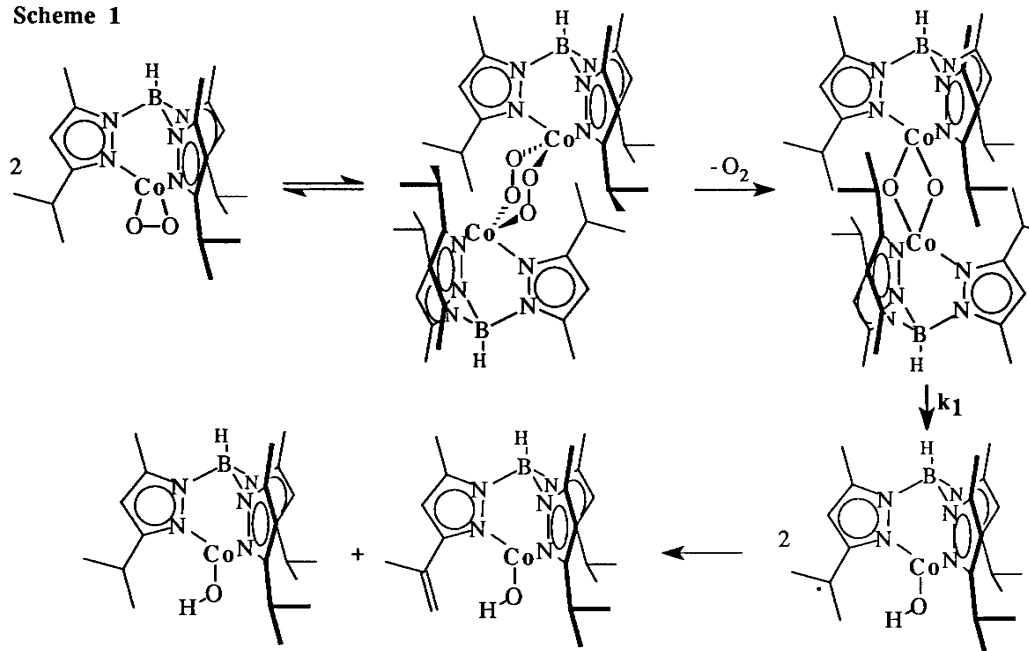


In order to lay the foundation for the design of catalytic cycles for the air oxidation of organic molecules, we are exploring both kinds of processes. The following paragraphs describe some results of relevant studies using cobalt and chromium complexes.

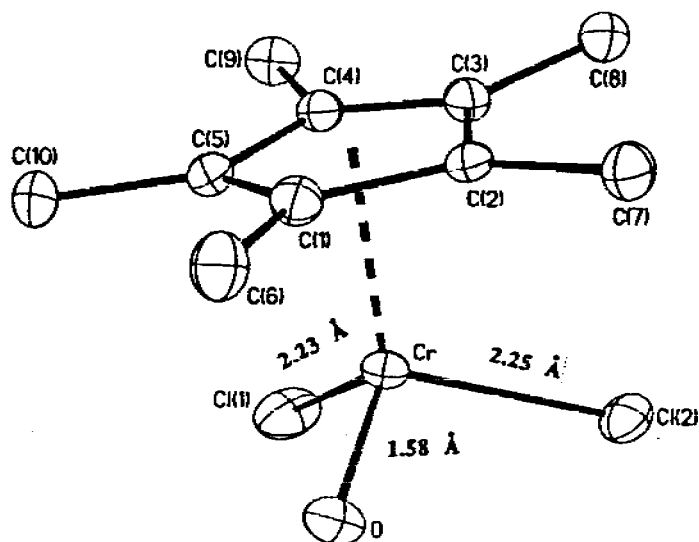
RESULTS

We are utilizing sterically encumbered tris(pyrazolyl)borate cobalt complexes in an attempt to create an artificial oxidation catalyst along the lines of equation 1. The first step was the preparation of the dioxygen complex $\text{Tp}^{\text{t-Bu,Me}}\text{Co}(\text{O}_2)$, the first example of a side-on bound superoxide complex.¹ $\text{Tp}^{\text{t-Bu,Me}}\text{Co}(\text{O}_2)$, while thermally stable by itself, was found to react with a second equivalent of metal - in the form of $\text{Tp}^{\text{t-Bu,Me}}\text{Co}(\text{N}_2)$ - to yield $\text{Tp}^{\text{t-Bu,Me}}\text{Co}-\text{OH}$, the ostensible product of O-O cleavage and hydrogen atom abstraction. As a test of this mechanistic hypothesis, the analogous $\text{Tp}^{\text{i-Pr,Me}}\text{Co}$ -system was prepared. $\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\text{O}_2)$ decomposes at ambient temperature, yielding the binuclear hydroxide, in which hydrogen has been abstracted from the isopropyl-substituents of the ligand. However, the reaction proceeds through a spectroscopically observable intermediate; based on several lines of evidence this intermediate has been assigned the formula $\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\mu\text{-O})_2\text{CoTp}^{\text{i-Pr,Me}}$. The decomposition of this complex involves hydrogen abstraction from the ligand; it exhibits an unusually large kinetic isotopic effect, which has been rationalized by invoking quantum mechanical tunneling.² The surprising difference in the thermal stabilities of $\text{Tp}^{\text{t-Bu,Me}}\text{Co}(\text{O}_2)$ and $\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\text{O}_2)$ has been traced to the accessibility of a dimer of the latter, i.e. $[\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\text{O}_2)]_2$, which can lose O₂ to form the dinuclear peroxo-complex.³ The whole sequence of transformations is shown in Scheme 1 below.

Scheme 1



We have used density functional calculations to determine the structures of reaction intermediates, and to estimate bond energies. Thus, our structural assignment of $\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\mu\text{-O})_2\text{CoTp}^{\text{i-Pr,Me}}$ is based on calculations, although the crystal structure of a similar molecule has recently been reported. The dissociation of formula $\text{Tp}^{\text{i-Pr,Me}}\text{Co}(\mu\text{-O})_2\text{CoTp}^{\text{i-Pr,Me}}$ into two mono-nuclear cobalt oxo complexes is energetically unfavorable.



Another system that accomplishes the activation of molecular dioxygen is the chromium complex $[\text{Cp}^*\text{Cr}(\mu\text{-Cl})(\text{Cl})_2]$. Upon exposure to O_2 gas, the latter rapidly and quantitatively yields $\text{Cp}^*\text{Cr}(\text{O})\text{Cl}_2$, a high-valent organometallic chromium(V) oxo complex. This molecule has been fully characterized, including a crystal structure determination, the result of which is shown in Figure 1.

Figure 1. The molecular structure of $\text{Cp}^*\text{Cr}(\text{O})\text{Cl}_2$ (1)

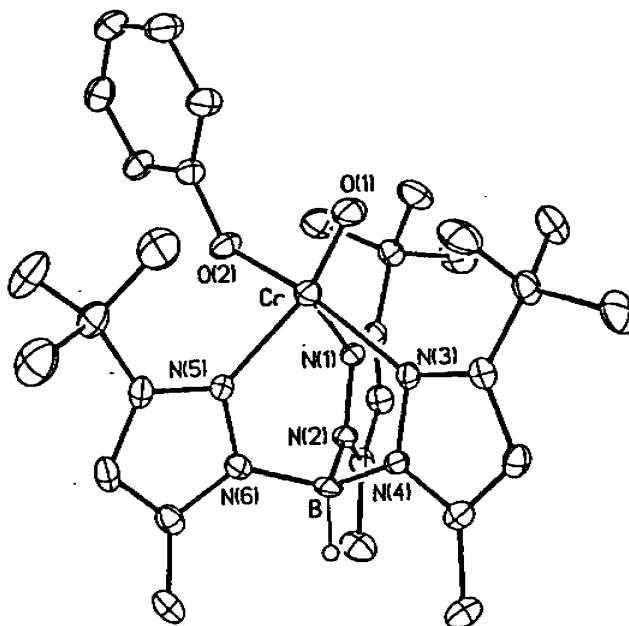
In light of its easy accessibility and the intense interest in catalytic processes utilizing O_2 as terminal oxidant, we have investigated the reactions of $\text{Cp}^*\text{Cr}(\text{O})\text{Cl}_2$ with oxidizable substrates. $\text{Cp}^*\text{Cr}(\text{O})\text{Cl}_2$ is capable of transferring oxygen atoms to easily oxidizable molecules (e.g. phosphines, arsines, sulfoxides), and it can activate weak X-H (X = C, N) bonds and dehydrogenate selected organic molecules (H_2O being the byproduct). Some of these reactions can be run catalytically in the presence of excess O_2 , e.g. the oxidation of triphenylphosphine and the dehydrogenation of 1,4-cyclohexadiene. However, this Cr^{V} oxo complex fails to transfer oxygen to less electron rich substrates (e.g. olefins, amines, CO), and it does not dehydrogenate or oxygenate common hydrocarbons.

The epoxidation of olefins by metal oxo complexes is a well preceded reaction, but efficient catalysts for the 'direct oxidation' (i.e. the code for 'epoxidation using O_2 as a reagent') of alkyl-substituted olefins (e.g. propene) are not known. The major problem of this transformation is the susceptibility of allylic C-H bonds to radical attack, providing competing reaction/decomposition pathways. $\text{Cp}^*\text{Cr}(\text{O})\text{Cl}_2$ does not react with olefins, such as norbornene, ethylene or cyclohexene. However, the expected products of these reactions, i.e. epoxide

complexes of the type $\text{Cp}^*\text{Cr}(\text{OC}_2\text{R}_4)\text{Cl}_2$ ($\text{R} = \text{H}$, alkyl) can be formed by reaction of $[\text{Cp}^*\text{CrCl}_2]_2$ with the appropriate epoxide. Thus it was not clear whether the oxygen atom transfer reaction fails for thermodynamic or kinetic reasons. This question has been addressed by DFT calculations, however. The calculations revealed that the oxygen atom transfer from the chromium complex to ethylene is thermodynamically favorable ($\Delta G \sim -17$ kcal), but faces a substantial activation barrier. Further research will be aimed at modifying the chromium complex and thereby decreasing the activation energy for the epoxidation.

Representative of the approach outlined in equation 2 is our recent study of the reaction of $\text{Tp}^{\text{t-Bu,Me}}\text{Cr-Ph}$ with O_2 .⁴ The product of this reaction is $\text{Tp}^{\text{t-Bu,Me}}\text{Cr}^{\text{IV}}(\text{O})\text{OPh}$ (see Fig. 2), i.e. the product of a formal insertion of oxygen into the metal-carbon bond. While O_2 insertion into metal alkyls are preceded, they are often thought to proceed by radical chain mechanisms. Mechanistic investigations showed, however, that this reaction is intramolecular (no crossover!) and proceeds via an IR-spectroscopically observable O_2 complex, i.e. $\text{Tp}^{\text{t-Bu,Me}}\text{Cr}(\text{O}_2)\text{Ph}$. The insertion of oxygen into metal-carbon bonds is of interest as a fundamental step in metal catalyzed oxidations of hydrocarbons; thus the transformation noted above is a possible step in a chromium mediated direct oxidation of benzene to phenol, a reaction of great commercial interest.

Figure 2. The molecular structure of $\text{Tp}^{\text{t-Bu,Me}}\text{Cr}(\text{O})\text{OPh}$; $\text{Cr-O}(1)$, 1.58Å; $\text{Cr-O}(2)$, 1.84Å.



SUMMARY

We have shown that various metal complexes can be used to activate dioxygen, i. e. to coordinate it and split it into reactive metal oxo moieties. The oxygenation of preformed metal carbon bonds can be accomplished by reaction of organometallics with O_2 , and we have demonstrated that the mechanism of O_2 -insertion does not have to be a radical chain process. Oxidation catalysis using O_2 (air) as a reagent remains an attractive option from an environmental standpoint. Our studies represent a contribution toward the fundamental understanding of the relevant chemistry.

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DEVELOPMENT OF HETEROGENEOUS ALDOL CONDENSATION CATALYSTS FOR IMPROVED ENVIRONMENTAL PERFORMANCE

James J. Spivey^{*1}, Ben W-L. Jang¹, Ramesh Subramanian^{1,4}, Apostolos A. Nikolopoulos¹, David Olsen², Tom Devon³ and Robert D. Culp³

¹Research Triangle Institute, P.O. Box 12194, RTP, NC 27709-2194

²Eastman Chemical Company, P.O. Box 1972, Kingsport, TN 37662-5150

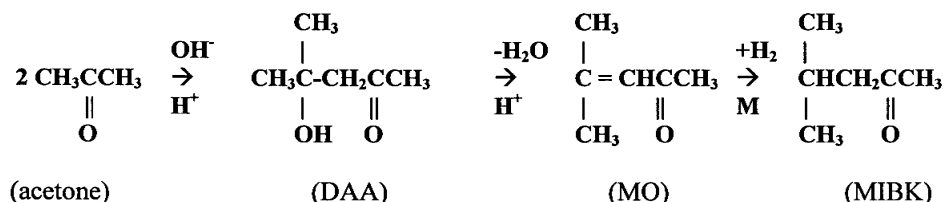
³Eastman Chemical Company, P.O. Box 7444, Longview, TX 75607-7444

⁴Present address: Johnson Matthey, West Deptford, NJ 08066

*Phone: (919) 541-8030; fax: (919) 541-8000; e-mail: jjs@rti.org

Introduction

The liquid-phase condensation of acetone is an important industrial process for the synthesis of a series of commodity chemicals like diacetone alcohol (DAA), mesityl oxide (MO) and methyl isobutyl ketone (MIBK)¹:



Aldol condensation reactions (and acetone condensation in particular) are practiced industrially using homogeneous liquid-based catalysts, like sodium and calcium hydroxide. Such processes generate wastewater streams that have to be neutralized and properly disposed, accounting for a significant portion of the selling price. Therefore, the development of suitable heterogeneous catalysts for aldol condensation would make this process more economical and environmentally benign.

Objective

The objective of this research is to obtain a better understanding of the condensation of acetone on heterogeneous catalysts. The functionality of these catalytic materials would have to be rather complex: the acetone condensation to DAA is catalyzed by either basic or acidic sites, the dehydration of DAA to MO is acid-catalyzed, and the hydrogenation of MO to MIBK would require appropriate metal sites^{2,3}.

Experimental

The acetone condensation to MIBK was studied on various *ex-situ* reduced metal-supported catalysts, the metal being Ni, Pd, or Pt, on various supports (Al_2O_3 , Nb_2O_5 , ZrP, ZSM-5, hydrotalcite). Catalyst characterization included external surface area measurements (BET), actual metal loading (ICP/OES) and metal dispersion (CO and H_2 pulse chemisorption), as well as acidity and basicity of the synthesized catalysts (NH_3 and CO_2 thermoprogrammed desorption, respectively). The reaction was performed in a liquid phase batch micro-reactor, typically at 118°C and 350 or 400 psig for 4-6 hours. Reaction products were analyzed using GC-FID and a capillary column (HP-1 methyl siloxane) for separation of compounds.

Results and Discussion

Reaction experiments showed that among the various metal and support combinations examined, the Pd- and Pt-supported HTC catalysts gave the highest selectivity to MIBK. Results obtained from a series of Pd/HTC and Pt/HTC samples of various metal loadings indicated that the 0.1% Pd/HTC catalyst gave the highest MIBK yield (31%), implying that minimal hydrogenation activity is required for the MO-to-MIBK reaction. The HTC support apparently catalyzes the condensation of acetone to MO, whereas the noble metal hydrogenates MO to MIBK. A reaction temperature increase favored the formation of MIBK and moderately shifted the product selectivity towards heavier (C_9+) condensation products. Selection of the appropriate reaction conditions may be critical for maximizing the activity for MIBK formation.

Catalyst	Conversion (mol%)	Selectivity (mol%)						
		DAA	MO	MIBK	IPA	DMH	DIBK	HGL
No catalyst	3.1	0	0	0	>99.9	0	0	0
HTC	19.6	14.4	84.9	0.6	0	0	0	0
0.1% Pd/HTC	38.0	14.3	0	82.2	0.6	1.2	1.8	0
0.5% Pd/HTC	29.7	11.8	0	71.7	14.7	0.6	1.0	0
1.5% Pd/HTC	28.2	9.0	0	61.8	27.7	0.5	0.8	0.2
0.1% Pt/HTC	34.0	12.0	0	52.4	32.6	0.4	0.8	0
0.5% Pt/HTC	32.7	6.0	0	38.1	52.5	0.3	0.5	0
1.5% Pt/HTC	30.8	6.6	0	33.6	56.8	0.2	0.3	0.3

DAA: diacetone alcohol (4-hydroxy-4-methyl-2-pentanone)

MO: mesityl oxide (4-methyl-3-penten-2-one)

MIBK: methyl isobutyl ketone (4-methyl-2-pentanone)

IPA: isopropanol (2-propanol)

DMH: dimethylheptanone(4,6-dimethyl-2-heptanone)
(2-methyl-2,4-pentanediol)

HGL: hexylene

DIBK: diisobutyl ketone (2,6-dimethyl-4-heptanone)

glycol

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3rd Annual Green Chemistry &
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Moving Toward Industrial Ecology

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**BIO-BASED SYNTHESIS
AND PROCESSING**

**SUCCESSFUL DEVELOPMENT OF HAZARD-FREE, USER-FRIENDLY GENETICALLY
ENGINEERED MICROORGANISMS FOR EFFECTIVE PRODUCTION OF
ENVIRONMENTALLY FRIENDLY CHEMICALS FROM RENEWABLE BIOMASS**

N. W. Y. Ho

Group Leader and Senior Research Scientist, Molecular Genetics Group,
Laboratory of Renewable Resources Engineering, Purdue University, West Lafayette, IN 47907

Microorganisms have been reliable tools for converting renewable biomass to useful products since the dawn of civilization. With the advances in genetic engineering technology, major alterations can be made on microorganisms resulting in widely broadening of the range of feedstocks that they can use and of the spectrum of products that they can produce. The challenge now is to use genetic engineering technology wisely and responsibly to program hazard-free, user-friendly microorganisms for effectively converting renewable biomass, particularly cellulosic biomass--an inexpensive, renewable resource abundantly available in the U.S. and in many other countries--to environmentally friendly industrial chemicals, especially those that can replace or substitute products traditionally produced through the petrochemical routes. In this paper, using the *Saccharomyces* yeasts as an example, we demonstrate that hazard-free, user-friendly microorganisms can successfully be genetically engineered by using recombinant DNA and gene cloning techniques either to use new feedstocks to produce products such as ethanol traditionally produced by the *Saccharomyces* yeasts, or to produce new products such as lactic acid from traditional feedstocks or new feedstocks.

Genetic Engineering of *Saccharomyces* Yeasts for Effective Conversion of Cellulosic Biomass to Ethanol as Renewable Environmentally Friendly Transportation Fuel

Numerous studies have proven that ethanol as a transportation fuel produces less air pollutants than gasoline. This environmentally friendly liquid fuel can be used directly as a neat fuel (100 %) or as a blend with gasoline at various concentrations. The raw material used for the production of ethanol fuel is renewable and abundantly available domestically. Thus, the use of ethanol to supplement or replace gasoline not only reduces air pollution and ensures a cleaner environment, but also reduces the dependency of our nation on imported foreign oil, protects our nation's energy security, and reduces our nation's trade deficit due to imported oil for the production of gasoline.

Ethanol has traditionally been produced by fermenting glucose-based food crops, such as cane sugar, corn starch, and other starch-rich grains, using yeasts, particularly *Saccharomyces* yeasts, which remain the only microorganisms used for large scale industrial ethanol production since the pre-industrial age. However, these agricultural crops are expensive and in limited supply.

Cellulosic biomass, which includes agriculture residues, waste streams from agricultural processing, sugarcane bagasse, municipal solid cellulosic wastes, yard and wood wastes, wastes from paper mills, etc. is an attractive feedstock for ethanol-fuel production by fermentation because cellulosic biomass is not only renewable and available domestically but also available at low cost and in great abundance. However, there are problems which must be solved for such ideal feedstocks to be economically converted to ethanol. One serious problem is that the major sugars derived from cellulosic biomass include not only glucose but also xylose with a ratio of glucose to xylose approximately 2 or 3 to 1 and that the *Saccharomyces* yeasts are unable to ferment xylose to ethanol or to use it for growth. It is generally agreed that unless both glucose and xylose from the cellulosic biomass could be fermented, the economics of converting cellulosic biomass to ethanol would not be favorable.

We demonstrated that with careful design and implementation, *Saccharomyces* yeasts were successfully genetically engineered not simply able to ferment xylose to ethanol but also able to (1) effectively direct the metabolic flux towards the production of ethanol rather than the production of byproducts such as xylitol, (2) effectively coferment both glucose and xylose simultaneously so that the mixed sugars will be fermented as fast as possible, (3) easily convert most *Saccharomyces* strains, particularly the superior glucose-fermenting industrial strains to coferment xylose in addition to glucose, and (4) use rich medium for growth and fermentation to make the engineered yeast grow and ferment sugars faster and to solve the potential waste problems by recycling used yeast cells for the production of crude yeast extracts for culturing new yeast cells. Furthermore, the final genetically engineered yeasts are stable and able to use either batch or continuous process for ethanol production without requiring the use of any special chemicals functioning as the selection pressure to maintain the cloned genes. This was accomplished (1) by constructing a functional metabolic pathway in yeast coupled by forcing the metabolic flux from xylose to the production of ethanol rather than to the production of the undesired

byproducts such as xylitol, (2) by overcoming the natural barrier "glucose effect" present in microorganisms so that the resulting engineered yeasts can coferment glucose and xylose simultaneously, (3) by designing an ideal system to select the yeast containing the cloned xylose-metabolizing genes and to maintain them in the engineered yeast, and (4) by developing a new method for effectively integrating multiple copies of multiple genes into the yeast chromosome(s).

Genetic Engineering of Saccharomyces Yeasts for the Production of New Products

Saccharomyces yeasts are hazard-free, user-friendly industrial microorganisms. With the development of recombinant DNA and gene cloning techniques, they can be genetically engineered to effectively produce numerous other products, particularly those, like ethanol, need to be produced in large scales.

Lactic acid is a chemical that has been used in the production of numerous industrial products such as cosmetic and pharmaceutical products. Recently, lactic acid has become the desired feedstock for the production of affordable biodegradable plastics, and the demand for such production should increase greatly for many years to come.

Currently the favorable industrial process for the production of lactic acid is by fermentation of glucose with lactic acid bacteria, which however is not a user-friendly microorganism for the production of lactic acid because the latter bacteria are strongly inhibited by the presence of lactic acid in the media, and therefore it is not an effective process for large scale production of lactic acid to be used for the production of biodegradable plastics. Logically, Saccharomyces yeasts can be genetically engineered for the effective production of lactic acid. This can be accomplished by the development of yeasts that are unable to produce ethanol, followed by cloning and overexpression of gene(s) responsible for the effective production of lactic acid in the latter yeasts. Our preliminary results indicated that such genetically engineered yeasts can be very effective for lactic acid production.

With continuing improvement of genetic manipulation of genes and gene cloning techniques, it is reasonable to assume that in the near future one can clone and effectively express any gene into any microorganism. Thus, it is the responsibility of scientists to genetically engineer hazard-free, user-friendly microorganisms for the effective production of safe and environmentally friendly industrial products.

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AFFORDABLE GENETICALLY ENGINEERED SOY BASED COMPOSITES, RESINS AND ADHESIVES

Richard P. Wool

Department of Chemical Engineering and Center for Composite Materials,
University of Delaware, Newark, DE 19716-3144

SUMMARY

Recently developed new soy-based plastics and adhesive materials are being evaluated for high volume applications in Agricultural Equipment (tractors and farming machines), Automotive (car and truck parts), Civil (bridges and highway components), Marine (pipes and offshore equipment), Rail Infrastructure (carriages, box cars and grain hoppers), Defense (Composite Armor) and the Construction Industry (formaldehyde-free particle board, ceilings, engineered lumber). Recent advances in genetic engineering, triglyceride chemical modification, natural fiber development and composite science offer significant opportunities for new improved materials from renewable resources with enhanced support for global sustainability.

SYNTHESIS AND MANUFACTURING OF SOY-BASED RESINS

The Affordable Composites from Renewable Resources (ACRES) group at the University of Delaware examined several hundred chemical pathways for the conversion of normal and genetically engineered soyoil triglycerides to high performance, inexpensive composites and plastics¹⁻³. The chemical modification of the oil was done with regard to cost, manufacturability, method of cure and final material properties (Patents Filed). The detailed

chemical pathways will be presented at the Green Chemistry conference and consist of both free radical and condensation reactions following suitable chemical functionalization of the natural oils. The first composites from soybeans were made by Henry Ford (Figure 1) about 50 years ago using protein macromolecules as the plastic resin. Much work has since been done on the plastic properties of high viscosity protein isolates. However, the synthesis of plastic materials first begins with low molecular weight, low viscosity monomers which are purified

and then converted to high molecular weight polymers having linear, branched and crosslinked structures. This is the approach taken by the ACRES group whereby low viscosity, chemically modified plant-based triglyceride oils are used as the starting monomers and we chemically control their properties to obtain a broad range of plastic, adhesive and composite materials. Genetic engineering serves to control/narrow the triglyceride fatty acid distribution function.



Figure 1. Henry Ford (1938) is shown testing the impact resistance of the first soy-based composite made from soy protein based plastic reinforced with glass. He grew many varieties of soybeans near the Detroit Auto Factory to evaluate optimal plant/material properties, the equivalent of today's genetically engineered plants used by the ACRES group at Delaware (Courtesy Henry Ford Museum, Michigan).

Four types of triglycerides typically make up a composite resin system, namely (a) Sizing, (b) Matrix, (c) Rubber Toughening, and (d) Material Modification. For natural fibers, or unsized glass or carbon, about 1% of the system consists of the sizing molecules. These have groups which allow them to bond both to the surface as well as to the matrix. A strong fiber-matrix interface bond is critical for high strength composites. The ability to apply the sizing insitu, offers considerable savings of time and cost, especially for all natural composites which are intended to be low cost.

The matrix consists of the dominant phase binding the fibers together in the composite and can be selected with respect to required material properties (hydrophilicity, biodegradability, flammability, dielectric, etc.). The rubber generating molecules (5-20%) can be made in situ or preformed, depending on the manufacturing conditions. The rubber particles when used at the optimal concentration, impart considerable impact resistance to both the neat resin and the composite. Other triglycerides are chemically modified to tailor the optical, thermal, electrical and mechanical properties of the composite.

RESULTS AND APPLICATIONS

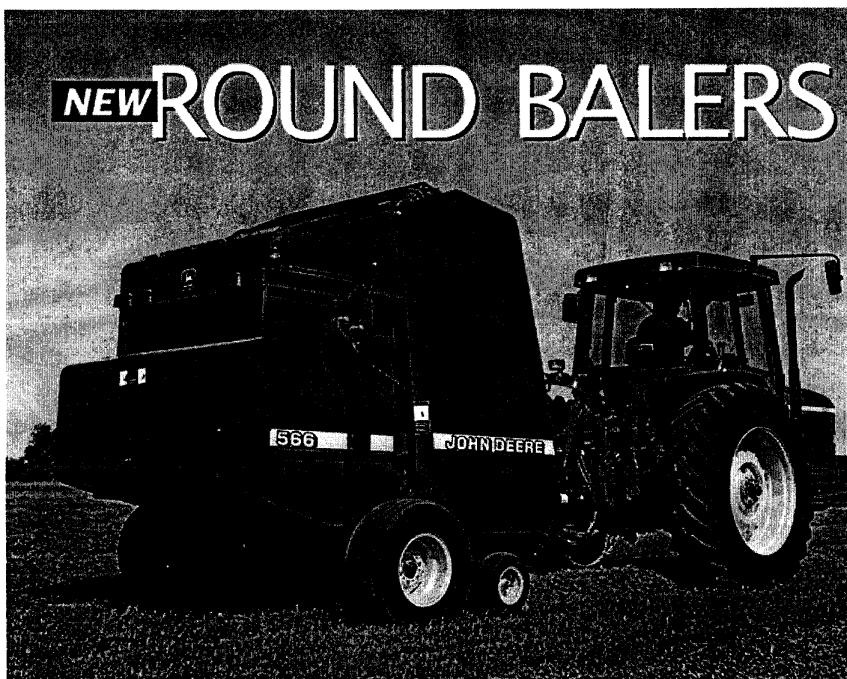
About a dozen new resin systems were derived from chemically modified triglycerides which gave excellent properties, and at a cost which is a fraction of current resin prices^{2,3}. The soy-based resins, when injected into molds and reacted by free radical or condensation mechanisms, produced high modulus, high T_g , thermosetting composites matrices. An example of a new commercial product manufactured from the soy-based resins is shown in Figure 2.

Typical properties of E-glass fiber filled resins (50% vol) gave tensile strengths of about 400 MPa (ASTM 3039-76), Young's Modulus of about 20 GPa and a fracture energy of about 50 kJ/m², which compare favorably with similar vinylester and polyester high performance composites (S. Khot, G. Palmese and R. P. Wool). Excellent resistance to acid, alkali, hot and cold water were found for some of these high performance ACRES composites. Others were found to swell in hot water in a manner dependent on the amount of hydrophilic groups contained in the triglyceride structure. The durability of the ACRES resins and composites is being field tested in tractor (hood and underhood) and ag-vehicle body components fabricated by RTM, APM and Hand-Lay-Up processes.

MODEL OIL STUDIES

A series of maleinized hydroxylated triglycerides were synthesized from model compounds and genetically engineered oils (supplied by DuPont) (J. LaScala and R. P. Wool). The object of this study was to determine the role of C=C unsaturation and triglyceride structure on the thermal and mechanical properties of the neat resin. The results indicate that significant improvements of viscosity, mechanical and thermal properties can be obtained with control of the triglyceride structure and functionality.

Figure 2. Round Hay Bailer. The 8x3' Panel containing the name "John Deere" was made from the ACRES soy-based resin (Courtesy, John Deere, Moline, Illinois).



This work is in progress using genetically engineered oils and model compounds with a focus on both rigid and soft plastic composites (J. LaScala, S. Bunker and R. P. Wool).

BIODEGRADABLE RESINS

Most of the composite resins were found to be non-biodegradable, as required for the traditional long-life applications, but some were found to be biodegradable through the action of lipase secreting enzymes, as would be expected from the structure of the natural oils. At least one resin exhibited rapid biodegradation in a soil burial test. The new soy-based biodegradable plastics are useful in applications where the biodegradability is an important component of the materials performance in aquatic and terrestrial environments, or in municipal solid waste management where composting and landfill reclamation are desirable.

ALL NATURAL COMPOSITES

Excellent inexpensive composites were made using natural fibers, such as hemp, straw, flax, and wood (G. Williams and R. P. Wool, J. Applied Composites, 1999). The soy-based resins can have a strong affinity for natural fibers and form an excellent fiber-matrix interface, as determined by SEM analysis of fractured composites. These composites are considered the most affordable high performance composites available and cost fractions of a dollar per kilo. They also form excellent inexpensive hybrid composites when used with mixtures of glass or carbon fibers (S. Morye, G. Williams and R. P. Wool). The use of natural fibers with these compatible resins allows a double environmental impact of replacing wood from trees with a more renewable resource such as flax or hemp, and petroleum oil with soyoil. Additionally, the new all-natural composites offer several alternatives for recycling and disposal of composites.

The bio-based resins can be significantly improved by genetic engineering of the triglyceride distribution with control of the structure and chemical modification. In addition to rigid composites, these materials are excellent for softer materials such as Pressure Sensitive Adhesives (S. Bunker and R. P. Wool), Rubbers with controlled cross-link density, Solid Rocket Propellant Binders, Composite Armored Vehicles (S. Naik and R.P. Wool) and Smart Adhesive that will adhere to most materials.

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DIRECT STARCH CONVERSION TO INDUSTRIAL BIOCATALYSTS USING TRANSGENIC GUNGAL SYSTEM

J. Gao

(Tel: 509-375-6639, jw.gao@pnl.gov)

B. S. Hooker, R. S. Skeen and D. B. Anderson

Bioprocessing Group, Pacific Northwest National Laboratory, Battelle Boulevard,
P.O. Box 999, MS K2-10, Richland, WA 99352

SUMMARY

The production of a bacterial enzyme, beta-glucuronidase (GUS), was investigated using a genetically modified starch-degrading *Saccharomyces* strain in suspension cultures of various waste starch sources. Different sources of starches including corn and waste potato starch were used for yeast biomass accumulation and GUS expression studies. In addition, a thermostable bacterial cellulase, *Acidothermus cellulolyticus* E1 endoglucanase was cloned into the plasmid expression vector and expressed in the starch-degrading *Saccharomyces* strain.

INTRODUCTION

Vast quantities of biomass are produced in the U.S. by the agricultural and food-processing industries, with only a small portion directly utilized for food, fiber, chemicals, or energy. A large fraction of the remaining biomass provides only marginal economic benefit and much of it must be managed as waste, incurring expenses to the producer or processor. For example, U.S. fruit and vegetable processors currently produce approximately 7.4 million dry metric tons of low-value byproduct (much of it sold at or near cost as livestock feed), and treat more than 300 million cubic meters of high BOD waste waters each year. Both of these byproduct streams are usually rich in starch, which could serve as an inexpensive yet robust feedstock for biotechnological processes for useful industrial enzyme production.

Natural yeast strains have been identified that can use starch as a primary growth substrate via complete or partial enzymatic hydrolysis^{1,2}. These yeast strains include *Saccharomycopsis fibuligera*, *Schwanniomyces castellii*, and *Saccharomyces diastaticus*^{1,2,3}. A fusion yeast cell strain of *Saccharomyces diastaticus* and *Saccharomyces cerevisiae* has also been demonstrated to degrade 60% of starch present in culture media within two days⁴. In addition, other natural *Saccharomyces* species can ferment starch and dextrin to ethanol⁵. For the past two decades, the starch-biotransforming yeast technology was mostly used for producing single cell protein as a cattle feed supplement^{2,3}. Others use this technology to improve ethanol production from starch and higher sugars^{6,7}. In the present study, a starch-degrading strain was genetically modified for the expression of a bacterial enzyme, beta-GUS and a thermostable enzyme, endoglucanase to demonstrate that this system may serve as means to produce inexpensive transgenic protein products.

MATERIALS AND METHODS

Escherichia coli strain Top 10' (Invitrogen Inc., Carlsbad, CA) was used as a host for routine cloning experiments. The yeast strain used in the study was a *Saccharomyces* strain provided by Dr. James R. Mattoon of University of Colorado, Colorado Springs, CO. Plasmid DNA purification, plasmid construction and transformation, and polymerase chain reaction (PCR) were conducted under standard molecular cloning techniques⁸. An EasySelect Expression Kit (Invitrogen) was used for preparing competent yeast cells for expression vector transformation. Upon transformation, the transformed yeast cells were plated onto a selective YPD agar plate containing 1.0% glucose, 0.5% yeast extract, 1.0% peptone, and 200 mg/l antibiotic Zeocin. After a 3-day incubation at 30°C, transformed yeast colonies were obtained.

The culture medium used for yeast biomass accumulation contains waste potato starch (Lamb-Weston, Richland, WA) or corn starch (Sigma, St. Louis, MO) as the primary carbon source supplemented with 1.0% peptone and 0.5% yeast extract as the nitrogen source. The expression medium contains 2% galactose, 1.0% peptone, and 0.5% yeast extract. All suspension cultures were grown aerobically at 30°C on an orbital shaker shaking at 200 rpm. Intracellular protein of transformed yeast biomass was extracted using the glass-bead disintegrating method in an extraction buffer containing 50 mM pH 7.0 sodium phosphate, 1 mM EDTA, 1 mM phenylmethylsulfonyl fluoride (PMSF), 10 mM beta-mercaptoethanol, and 0.1% triton X-100. The extracted protein samples were assayed for protein concentration using the Bio-Rad protein assay (Bio-Rad Laboratories, Hercules, CA).

GUS activity was assayed using an enzymatic reaction in which one unit of GUS activity is defined as the amount of GUS that produces one pmole 4-methylumbelliferone (MU) from 4-methylumbelliferyl-beta-D-glucuronide (MUG) per minute at 37 °C. E1 Endoglucanase activity was assayed using an enzymatic reaction in which one unit of GUS activity is defined as the amount of E1 endoglucanase that produces one pmole MU from 4-methylumbelliferyl-beta-D-cellobioside (MUC) per minute at 55°C. The MU fluorescence was assayed in a DyNA QUANT 200 fluorometer (Pharmacia Biotech, Piscataway, NJ). The specific activity calculated as the units of activity per milligram of total protein.

RESULTS AND DISCUSSIONS

Plasmid vectors were constructed to effectively transform the starch-degrading *Saccharomyces* strain. A plasmid vector pGA2026 (4,961 bp) was constructed by replacing the ampicillin resistance and URA3 genes in a plasmid vector pYES (Invitrogen) with the antibiotic Zeocin resistance gene of the plasmid vector pGAPZ α -A (Invitrogen). The plasmid pGA2026 enables the selection of transformed *Saccharomyces* strains without using uracil-deficient selection medium. The plasmid pGA2026 contains an expression cassette for foreign gene expression under the control of an inducible galactokinase promoter (GAL1), a T7 RNA promoter and a T_{cyc1} terminator. In addition, the plasmid also contains a 2 μ m DNA fragment for plasmid replication in *Saccharomyces* strains, a Co1E1 origin for plasmid replication during gene manipulation in *E. coli* strains, a f1 phage origin, and the antibiotic Zeocin resistance gene for both yeast and *E. coli* selection during gene manipulation after transformation. A bacterial *gus* gene⁹ was subsequently cloned into pGA2026 to form GUS expression vector pGA2028. In addition, the starch-degrading yeast strain *Saccharomyces* was also used to express a thermostable cellulolytic enzyme gene, endoglucanase from *Acidothermus cellulolyticus*¹⁰. The beta-1,4-endoglucanase (E1) precursor gene was obtained from Steven R. Thomas of the National Renewable Energy Laboratory in Golden, CO. The E1 gene was cloned out and ligated into vector pGA2026 to form the E1 expression vector pGA2035.

Upon pGA2028 vector transformation, ten transformed yeast colonies were streak-purified on fresh selective YPD agar plates and single colonies were used in batch cultures for GUS expression. The yeast colonies were first grown aerobically in YPD medium for 16 hours. The propagated biomass was transferred into expression medium containing promoter activity inducer, galactose, for GUS expression. After 5 hours, yeast biomass was harvested and intracellular protein was extracted. Table 1 illustrates the results of GUS specific activities of ten

Table 1. Transgenic GUS activity in trans- formed starch degrading

Cone No.	GUS Specific Activity (unit/mg)
Control ^a	16
1	3,469
2	10,057
3	4,061
4	7,013
5	5,309
6	7,786
7	6,035
8	4,439
9	4,000
10	8,346

^aNo genetic transformation

different transformed clones, indicating that GUS can be highly expressed in the transgenic yeast host under the control of GAL1 promoter. The highest specific activity obtained in the culture was 10,057 units per mg of extracted intracellular protein. There was no extracellular GUS activity found since there was no secretion transit peptide sequence to the *gus* gene.

Starch medium was also used to cultivate transformed yeast clones for GUS expression. Corn and waste potato starches were used. The culture medium is a sugar-free medium and contains 1.0% corn or potato starch supplemented with 1.0% peptone and 0.5% yeast extract. After a 2-day growth period in the starch medium, the biomass was collected and GUS was induced in a production medium primarily containing 2% galactose as inducer. Cells were harvested periodically and intracellular protein was extracted. Results indicate that GUS activity reached 13,396 units/mg of extracted intracellular protein after 4-hour induction. The GUS activities leveled off thereafter, indicating stable expression of GUS in the cultures.

E1 endoglucanase expression was conducted by transforming the starch-degrading yeast strain *Saccharomyces* by vector pGA2035. Upon transformation, the transformed yeast colonies were obtained on selective YPD agar media containing antibiotic Zeocin after 3-day incubation at 30°C. Twelve transformed yeast colonies were streak-purified on fresh selective YPD agar plates and single colonies were used in batch cultures for E1 endoglucanase expression. The yeast colonies were first grown aerobically in YPD medium for 16 hours. The propagated biomass was transferred into expression medium containing the promoter activity inducer, galactose, for E1 endoglucanase expression. After a 5-hour inducing period, yeast biomass was harvested and intracellular protein was extracted in the extraction buffer using the glass-bead disintegrating method. Table 2 shows the results of E1 endoglucanase specific activities of twelve different transformed clones. The highest specific activity obtained in the culture was 1,724 units per mg of extracted intracellular protein. However, there was no secreted E1 endoglucanase detected in the culture medium even though a native leader sequence was used in the E1 expression construct. This is probably due to the strong binding activity of the E1 cellulose-binding domain to the cell wall cellulose, inhibiting E1 secretion. Preliminary results showed that the removal of the cellulose-binding domain greatly enhanced E1 enzyme secretion.

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Table 2. Transgenic thermostable E1 endoglucanase activity in transformed starch degrading *Saccharomyces*.

Clone No.	E1 endoglucanase Specific Activity (unit/mg)
Control ^a	7
1	1,724
2	961
3	1,241
4	1,383
5	1,294
6	1,273
7	1,257
8	1,111
9	1,501
10	1,258
11	508
12	1,476

^aNo genetic transformation

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IN-VITRO ENZYME CATALYZED POLYMER SYNTHESIS: NATURAL CATALYSTS DOING NON-NATURAL THINGS

Richard A. Gross* and David L. Kaplan†

Center for Biocatalysis and Bioprocessing of Macromolecules

*Polytechnic University, Professor and Herman F. Mark Chair, Six Metrotech Center, Brooklyn, NY 11201
(rgross@poly.edu)

†Tufts University, Professor of Chemical Engineering, 4 Colby Street, Medford, MA 02155
(dkaplan1@tufts.edu)

Overview

Our program is focused on the use of enzymes in the synthesis and modification of polymers. Nature provides an extraordinarily diverse range of enzyme catalysts that carry out oxidations, reductions, esterification, amidation, glycolysis, decarboxylation, transamination and many other reactions. Our interests are in determining how to apply natural and engineered enzymes to transformations that result in monomer and polymer synthesis, polymer modification and controlled crosslinking reactions. An extraordinary attribute of enzymes is their ability to provide simple direct routes to products where enantio- or regioselective control is beneficial. Another benefit is the potential of enzyme transformations to provide lower energy pathways to products. Enzyme-catalysis can also circumvent the need for heavy metal catalysts that are environmentally unsafe.

The results of our work on polyester synthesis by lipase catalysis have shown that these methods will be very important in the generation polyesters with functional pendant groups without tedious protection-deprotection steps. *Exploitation of enzyme regioselectivity has also proved important for the preparation of novel block copolymer architectures.* For example, a versatile family of surface active agents have been prepared that consist of a carbohydrate head group attached regiospecifically to either an oligo(ester) or an oligo(carbonate) hydrophobic chain segment. In addition, this structural theme has been used to prepare new multi-arm heteroblock copolymers. Architectural control is an outcome of using natural structures as building blocks in combination with enzyme-catalyzed transformations. Similarly, we have exploited peroxidase and laccase reactions in free radical polymerizations. An outcome of these studies has been the ability to gain control of the structural features of the polymer products through a combination of monomer choice, solvent engineering and enzyme choice. An important component in current projects is the application of accelerated directed evolution along with the appropriate high-throughput assays to develop enzyme-catalysts that have improved catalytic efficiency.

Topics of presentation

1) Propagation kinetics from the ring-opening polymerizations of pentadecalactone (PDL), ϵ -caprolactone (ϵ -CL) and trimethylene carbonate were studied using the lipases from a *Pseudomonas* sp. (immobilized on Celite 521) and *Candida antarctica* (Novozym 435). The experimental results were compared to two living kinetic models describing slow-initiation and slow exchanges between dormant and active species. The use of an interferometric technique is providing new information on the distribution of enzymes and real-time monitoring of polymerizations within an immobilization matrix. Recent work using solvent engineering has resulted in large enhancements in enzyme stability at elevated temperatures and, as a result, rapid polymerization reactions. For example, using recently developed conditions for these polymerizations, it is now possible to reach high monomer conversions within a few minutes

2) Lipase-catalyzed ring-opening polymerization of lactones and carbonates using ethylglucopyranoside (EGP) as a multifunctional initiator were carried out. For example, products of variable oligo(-CL) chain length ($M_n = 450, 2200$ g/mol) were formed by variation of the -CL/EGP ratio. This family of amphiphilic structures was used to prepare heteromulti-arm block copolymers of controlled three dimensional structure.

3) Pendant glycolipids placed along polyacrylate chains are of interest as theological modifiers and as biactive agents that, for example, may function in immunomodulation. A new 6-O-acryl-sophorolipid monomer was prepared by a regioselective lipase-catalyzed acrylation of a sophorolipid macrolactone. The homopolymerization of the 6-O-acryl-sophorolipid derivative and its copolymerization with acrylic acid and acrylamide are reported.

Further exploration of *in-vitro* enzyme-catalysis will undoubtedly lead to a better understanding of the strengths and limitations of this approach. By integration of genetic methods to enhance catalytic activity, we hope to

overcome certain limitations and broaden the impact of these methods.

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3rd Annual Green Chemistry &
Engineering Conference

Moving Toward Industrial Ecology

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**BIO-BASED SYNTHESIS
AND PROCESSING**

SUCCESSFUL DEVELOPMENT OF HAZARD-FREE, USER-FRIENDLY GENETICALLY ENGINEERED MICROORGANISMS FOR EFFECTIVE PRODUCTION OF ENVIRONMENTALLY FRIENDLY CHEMICALS FROM RENEWABLE BIOMASS

N. W. Y. Ho

Group Leader and Senior Research Scientist, Molecular Genetics Group,
Laboratory of Renewable Resources Engineering, Purdue University, West Lafayette, IN 47907

Microorganisms have been reliable tools for converting renewable biomass to useful products since the dawn of civilization. With the advances in genetic engineering technology, major alterations can be made on microorganisms resulting in widely broadening of the range of feedstocks that they can use and of the spectrum of products that they can produce. The challenge now is to use genetic engineering technology wisely and responsibly to program hazard-free, user-friendly microorganisms for effectively converting renewable biomass, particularly cellulosic biomass--an inexpensive, renewable resource abundantly available in the U.S. and in many other countries--to environmentally friendly industrial chemicals, especially those that can replace or substitute products traditionally produced through the petrochemical routes. In this paper, using the *Saccharomyces* yeasts as an example, we demonstrate that hazard-free, user-friendly microorganisms can successfully be genetically engineered by using recombinant DNA and gene cloning techniques either to use new feedstocks to produce products such as ethanol traditionally produced by the *Saccharomyces* yeasts, or to produce new products such as lactic acid from traditional feedstocks or new feedstocks.

Genetic Engineering of *Saccharomyces* Yeasts for Effective Conversion of Cellulosic Biomass to Ethanol as Renewable Environmentally Friendly Transportation Fuel

Numerous studies have proven that ethanol as a transportation fuel produces less air pollutants than gasoline. This environmentally friendly liquid fuel can be used directly as a neat fuel (100 %) or as a blend with gasoline at various concentrations. The raw material used for the production of ethanol fuel is renewable and abundantly available domestically. Thus, the use of ethanol to supplement or replace gasoline not only reduces air pollution and ensures a cleaner environment, but also reduces the dependency of our nation on imported foreign oil, protects our nation's energy security, and reduces our nation's trade deficit due to imported oil for the production of gasoline.

Ethanol has traditionally been produced by fermenting glucose-based food crops, such as cane sugar, corn starch, and other starch-rich grains, using yeasts, particularly *Saccharomyces* yeasts, which remain the only microorganisms used for large scale industrial ethanol production since the pre-industrial age. However, these agricultural crops are expensive and in limited supply.

Cellulosic biomass, which includes agriculture residues, waste streams from agricultural processing, sugarcane bagasse, municipal solid cellulosic wastes, yard and wood wastes, wastes from paper mills, etc. is an attractive feedstock for ethanol-fuel production by fermentation because cellulosic biomass is not only renewable and available domestically but also available at low cost and in great abundance. However, there are problems which must be solved for such ideal feedstocks to be economically converted to ethanol. One serious problem is that the major sugars derived from cellulosic biomass include not only glucose but also xylose with a ratio of glucose to xylose approximately 2 or 3 to 1 and that the *Saccharomyces* yeasts are unable to ferment xylose to ethanol or to use it for growth. It is generally agreed that unless both glucose and xylose from the cellulosic biomass could be fermented, the economics of converting cellulosic biomass to ethanol would not be favorable.

We demonstrated that with careful design and implementation, *Saccharomyces* yeasts were successfully genetically engineered not simply able to ferment xylose to ethanol but also able to (1) effectively direct the metabolic flux towards the production of ethanol rather than the production of byproducts such as xylitol, (2) effectively coferment both glucose and xylose simultaneously so that the mixed sugars will be fermented as fast as possible, (3) easily convert most *Saccharomyces* strains, particularly the superior glucose-fermenting industrial strains to coferment xylose in addition to glucose, and (4) use rich medium for growth and fermentation to make the engineered yeast grow and ferment sugars faster and to solve the potential waste problems by recycling used yeast cells for the production of crude yeast extracts for culturing new yeast cells. Furthermore, the final genetically engineered yeasts are stable and able to use either batch or continuous process for ethanol production without requiring the use of any special chemicals functioning as the selection pressure to maintain the cloned genes. This was accomplished (1) by constructing a functional metabolic pathway in yeast coupled by forcing the metabolic flux from xylose to the production of ethanol rather than to the production of the undesired

byproducts such as xylitol, (2) by overcoming the natural barrier "glucose effect" present in microorganisms so that the resulting engineered yeasts can coferment glucose and xylose simultaneously, (3) by designing an ideal system to select the yeast containing the cloned xylose-metabolizing genes and to maintain them in the engineered yeast, and (4) by developing a new method for effectively integrating multiple copies of multiple genes into the yeast chromosome(s).

Genetic Engineering of Saccharomyces Yeasts for the Production of New Products

Saccharomyces yeasts are hazard-free, user-friendly industrial microorganisms. With the development of recombinant DNA and gene cloning techniques, they can be genetically engineered to effectively produce numerous other products, particularly those, like ethanol, need to be produced in large scales.

Lactic acid is a chemical that has been used in the production of numerous industrial products such as cosmetic and pharmaceutical products. Recently, lactic acid has become the desired feedstock for the production of affordable biodegradable plastics, and the demand for such production should increase greatly for many years to come.

Currently the favorable industrial process for the production of lactic acid is by fermentation of glucose with lactic acid bacteria, which however is not a user-friendly microorganism for the production of lactic acid because the latter bacteria are strongly inhibited by the presence of lactic acid in the media, and therefore it is not an effective process for large scale production of lactic acid to be used for the production of biodegradable plastics. Logically, Saccharomyces yeasts can be genetically engineered for the effective production of lactic acid. This can be accomplished by the development of yeasts that are unable to produce ethanol, followed by cloning and overexpression of gene(s) responsible for the effective production of lactic acid in the latter yeasts. Our preliminary results indicated that such genetically engineered yeasts can be very effective for lactic acid production.

With continuing improvement of genetic manipulation of genes and gene cloning techniques, it is reasonable to assume that in the near future one can clone and effectively express any gene into any microorganism. Thus, it is the responsibility of scientists to genetically engineer hazard-free, user-friendly microorganisms for the effective production of safe and environmentally friendly industrial products.

Acknowledgment

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AFFORDABLE GENETICALLY ENGINEERED SOY BASED COMPOSITES, RESINS AND ADHESIVES

Richard P. Wool

Department of Chemical Engineering and Center for Composite Materials,
University of Delaware, Newark, DE 19716-3144

SUMMARY

Recently developed new soy-based plastics and adhesive materials are being evaluated for high volume applications in Agricultural Equipment (tractors and farming machines), Automotive (car and truck parts), Civil (bridges and highway components), Marine (pipes and offshore equipment), Rail Infrastructure (carriages, box cars and grain hoppers), Defense (Composite Armor) and the Construction Industry (formaldehyde-free particle board, ceilings, engineered lumber). Recent advances in genetic engineering, triglyceride chemical modification, natural fiber development and composite science offer significant opportunities for new improved materials from renewable resources with enhanced support for global sustainability.

SYNTHESIS AND MANUFACTURING OF SOY-BASED RESINS

The Affordable Composites from Renewable Resources (ACRES) group at the University of Delaware examined several hundred chemical pathways for the conversion of normal and genetically engineered soy oil triglycerides to high performance, inexpensive composites and plastics¹⁻³. The chemical modification of the oil was done with regard to cost, manufacturability, method of cure and final material properties (Patents Filed). The detailed

chemical pathways will be presented at the Green Chemistry conference and consist of both free radical and condensation reactions following suitable chemical functionalization of the natural oils. The first composites from soybeans were made by Henry Ford (Figure 1) about 50 years ago using protein macromolecules as the plastic resin. Much work has since been done on the plastic properties of high viscosity protein isolates. However, the synthesis of plastic materials first begins with low molecular weight, low viscosity monomers which are purified

and then converted to high molecular weight polymers having linear, branched and crosslinked structures. This is the approach taken by the ACRES group whereby low viscosity, chemically modified plant-based triglyceride oils are used as the starting monomers and we chemically control their properties to obtain a broad range of plastic, adhesive and composite materials. Genetic engineering serves to control/narrow the triglyceride fatty acid distribution function.



Figure 1. Henry Ford (1938) is shown testing the impact resistance of the first soy-based composite made from soy protein based plastic reinforced with glass. He grew many varieties of soybeans near the Detroit Auto Factory to evaluate optimal plant/material properties, the equivalent of today's genetically engineered plants used by the ACRES group at Delaware (Courtesy Henry Ford Museum, Michigan).

Four types of triglycerides typically make up a composite resin system, namely (a) Sizing, (b) Matrix, (c) Rubber Toughening, and (d) Material Modification. For natural fibers, or unsized glass or carbon, about 1% of the system consists of the sizing molecules. These have groups which allow them to bond both to the surface as well as to the matrix. A strong fiber-matrix interface bond is critical for high strength composites. The ability to apply the sizing insitu, offers considerable savings of time and cost, especially for all natural composites which are intended to be low cost.

The matrix consists of the dominant phase binding the fibers together in the composite and can be selected with respect to required material properties (hydrophilicity, biodegradability, flammability, dielectric, etc.). The rubber generating molecules (5-20%) can be made in situ or preformed, depending on the manufacturing conditions. The rubber particles when used at the optimal concentration, impart considerable impact resistance to both the neat resin and the composite. Other triglycerides are chemically modified to tailor the optical, thermal, electrical and mechanical properties of the composite.

RESULTS AND APPLICATIONS

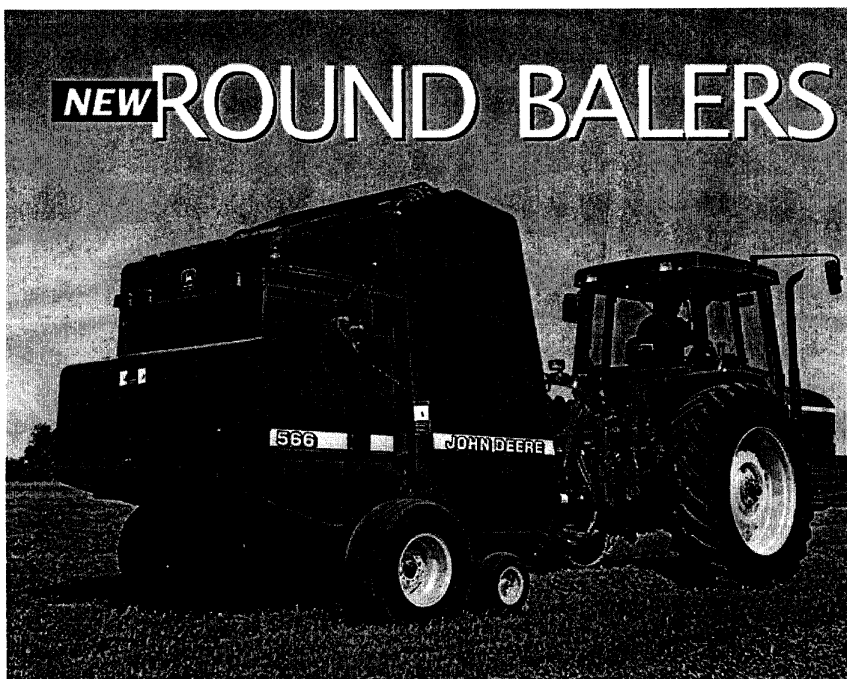
About a dozen new resin systems were derived from chemically modified triglycerides which gave excellent properties, and at a cost which is a fraction of current resin prices^{2,3}. The soy-based resins, when injected into molds and reacted by free radical or condensation mechanisms, produced high modulus, high T_g , thermosetting composites matrices. An example of a new commercial product manufactured from the soy-based resins is shown in Figure 2.

Typical properties of E-glass fiber filled resins (50% vol) gave tensile strengths of about 400 MPa (ASTM 3039-76), Young's Modulus of about 20 GPa and a fracture energy of about 50 kJ/m², which compare favorably with similar vinylester and polyester high performance composites (S. Khot, G. Palmese and R. P. Wool). Excellent resistance to acid, alkali, hot and cold water were found for some of these high performance ACRES composites. Others were found to swell in hot water in a manner dependent on the amount of hydrophilic groups contained in the triglyceride structure. The durability of the ACRES resins and composites is being field tested in tractor (hood and underhood) and ag-vehicle body components fabricated by RTM, APM and Hand-Lay-Up processes.

MODEL OIL STUDIES

A series of maleinized hydroxylated triglycerides were synthesized from model compounds and genetically engineered oils (supplied by DuPont) (J. LaScala and R. P. Wool). The object of this study was to determine the role of C=C unsaturation and triglyceride structure on the thermal and mechanical properties of the neat resin. The results indicate that significant improvements of viscosity, mechanical and thermal properties can be obtained with control of the triglyceride structure and functionality.

Figure 2. Round Hay Bailer. The 8x3' Panel containing the name "John Deere" was made from the ACRES soy-based resin (Courtesy, John Deere, Moline, Illinois).



This work is in progress using genetically engineered oils and model compounds with a focus on both rigid and soft plastic composites (J. LaScala, S. Bunker and R. P. Wool).

BIODEGRADABLE RESINS

Most of the composite resins were found to be non-biodegradable, as required for the traditional long-life applications, but some were found to be biodegradable through the action of lipase secreting enzymes, as would be expected from the structure of the natural oils. At least one resin exhibited rapid biodegradation in a soil burial test. The new soy-based biodegradable plastics are useful in applications where the biodegradability is an important component of the materials performance in aquatic and terrestrial environments, or in municipal solid waste management where composting and landfill reclamation are desirable.

ALL NATURAL COMPOSITES

Excellent inexpensive composites were made using natural fibers, such as hemp, straw, flax, and wood (G. Williams and R. P. Wool, J. Applied Composites, 1999). The soy-based resins can have a strong affinity for natural fibers and form an excellent fiber-matrix interface, as determined by SEM analysis of fractured composites. These composites are considered the most affordable high performance composites available and cost fractions of a dollar per kilo. They also form excellent inexpensive hybrid composites when used with mixtures of glass or carbon fibers (S. Morye, G. Williams and R. P. Wool). The use of natural fibers with these compatible resins allows a double environmental impact of replacing wood from trees with a more renewable resource such as flax or hemp, and petroleum oil with soyoil. Additionally, the new all-natural composites offer several alternatives for recycling and disposal of composites.

The bio-based resins can be significantly improved by genetic engineering of the triglyceride distribution with control of the structure and chemical modification. In addition to rigid composites, these materials are excellent for softer materials such as Pressure Sensitive Adhesives (S. Bunker and R. P. Wool), Rubbers with controlled cross-link density, Solid Rocket Propellant Binders, Composite Armored Vehicles (S. Naik and R.P. Wool) and Smart Adhesive that will adhere to most materials.

Acknowledgments

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DIRECT STARCH CONVERSION TO INDUSTRIAL BIOCATALYSTS USING TRANSGENIC GUNGAL SYSTEM

J. Gao

(Tel: 509-375-6639, jw.gao@pnl.gov)

B. S. Hooker, R. S. Skeen and D. B. Anderson

Bioprocessing Group, Pacific Northwest National Laboratory, Battelle Boulevard,
P.O. Box 999, MS K2-10, Richland, WA 99352

SUMMARY

The production of a bacterial enzyme, beta-glucuronidase (GUS), was investigated using a genetically modified starch-degrading *Saccharomyces* strain in suspension cultures of various waste starch sources. Different sources of starches including corn and waste potato starch were used for yeast biomass accumulation and GUS expression studies. In addition, a thermostable bacterial cellulase, *Acidothermus cellulolyticus* E1 endoglucanase was cloned into the plasmid expression vector and expressed in the starch-degrading *Saccharomyces* strain.

INTRODUCTION

Vast quantities of biomass are produced in the U.S. by the agricultural and food-processing industries, with only a small portion directly utilized for food, fiber, chemicals, or energy. A large fraction of the remaining biomass provides only marginal economic benefit and much of it must be managed as waste, incurring expenses to the producer or processor. For example, U.S. fruit and vegetable processors currently produce approximately 7.4 million dry metric tons of low-value byproduct (much of it sold at or near cost as livestock feed), and treat more than 300 million cubic meters of high BOD waste waters each year. Both of these byproduct streams are usually rich in starch, which could serve as an inexpensive yet robust feedstock for biotechnological processes for useful industrial enzyme production.

Natural yeast strains have been identified that can use starch as a primary growth substrate via complete or partial enzymatic hydrolysis^{1,2}. These yeast strains include *Saccharomycopsis fibuligera*, *Schwanniomyces castellii*, and *Saccharomyces diastaticus*^{1,2,3}. A fusion yeast cell strain of *Saccharomyces diastaticus* and *Saccharomyces cerevisiae* has also been demonstrated to degrade 60% of starch present in culture media within two days⁴. In addition, other natural *Saccharomyces* species can ferment starch and dextrin to ethanol⁵. For the past two decades, the starch-biotransforming yeast technology was mostly used for producing single cell protein as a cattle feed supplement^{2,3}. Others use this technology to improve ethanol production from starch and higher sugars^{6,7}. In the present study, a starch-degrading strain was genetically modified for the expression of a bacterial enzyme, beta-GUS and a thermostable enzyme, endoglucanase to demonstrate that this system may serve as means to produce inexpensive transgenic protein products.

MATERIALS AND METHODS

Escherichia coli strain Top 10' (Invitrogen Inc., Carlsbad, CA) was used as a host for routine cloning experiments. The yeast strain used in the study was a *Saccharomyces* strain provided by Dr. James R. Mattoon of University of Colorado, Colorado Springs, CO. Plasmid DNA purification, plasmid construction and transformation, and polymerase chain reaction (PCR) were conducted under standard molecular cloning techniques⁸. An EasySelect Expression Kit (Invitrogen) was used for preparing competent yeast cells for expression vector transformation. Upon transformation, the transformed yeast cells were plated onto a selective YPD agar plate containing 1.0% glucose, 0.5% yeast extract, 1.0% peptone, and 200 mg/l antibiotic Zeocin. After a 3-day incubation at 30°C, transformed yeast colonies were obtained.

The culture medium used for yeast biomass accumulation contains waste potato starch (Lamb-Weston, Richland, WA) or corn starch (Sigma, St. Louis, MO) as the primary carbon source supplemented with 1.0% peptone and 0.5% yeast extract as the nitrogen source. The expression medium contains 2% galactose, 1.0% peptone, and 0.5% yeast extract. All suspension cultures were grown aerobically at 30°C on an orbital shaker shaking at 200 rpm. Intracellular protein of transformed yeast biomass was extracted using the glass-bead disintegrating method in an extraction buffer containing 50 mM pH 7.0 sodium phosphate, 1 mM EDTA, 1 mM phenylmethylsulfonyl fluoride (PMSF), 10 mM beta-mercaptoethanol, and 0.1% triton X-100. The extracted protein samples were assayed for protein concentration using the Bio-Rad protein assay (Bio-Rad Laboratories, Hercules, CA).

GUS activity was assayed using an enzymatic reaction in which one unit of GUS activity is defined as the amount of GUS that produces one pmole 4-methylumbelliferone (MU) from 4-methylumbelliferyl-beta-D-glucuronide (MUG) per minute at 37 °C. E1 Endoglucanase activity was assayed using an enzymatic reaction in which one unit of GUS activity is defined as the amount of E1 endoglucanase that produces one pmole MU from 4-methylumbelliferyl-beta-D-cellobioside (MUC) per minute at 55°C. The MU fluorescence was assayed in a DyNA QUANT 200 fluorometer (Pharmacia Biotech, Piscataway, NJ). The specific activity calculated as the units of activity per milligram of total protein.

RESULTS AND DISCUSSIONS

Plasmid vectors were constructed to effectively transform the starch-degrading *Saccharomyces* strain. A plasmid vector pGA2026 (4,961 bp) was constructed by replacing the ampicillin resistance and URA3 genes in a plasmid vector pYES (Invitrogen) with the antibiotic Zeocin resistance gene of the plasmid vector pGAPZ α -A (Invitrogen). The plasmid pGA2026 enables the selection of transformed *Saccharomyces* strains without using uracil-deficient selection medium. The plasmid pGA2026 contains an expression cassette for foreign gene expression under the control of an inducible galactokinase promoter (GAL1), a T7 RNA promoter and a T_{cyc1} terminator. In addition, the plasmid also contains a 2 μ m DNA fragment for plasmid replication in *Saccharomyces* strains, a Co1E1 origin for plasmid replication during gene manipulation in *E. coli* strains, a f1 phage origin, and the antibiotic Zeocin resistance gene for both yeast and *E. coli* selection during gene manipulation after transformation. A bacterial *gus* gene⁹ was subsequently cloned into pGA2026 to form GUS expression vector pGA2028. In addition, the starch-degrading yeast strain *Saccharomyces* was also used to express a thermostable cellulolytic enzyme gene, endoglucanase from *Acidothermus cellulolyticus*¹⁰. The beta-1,4-endoglucanase (E1) precursor gene was obtained from Steven R. Thomas of the National Renewable Energy Laboratory in Golden, CO. The E1 gene was cloned out and ligated into vector pGA2026 to form the E1 expression vector pGA2035.

Upon pGA2028 vector transformation, ten transformed yeast colonies were streak-purified on fresh selective YPD agar plates and single colonies were used in batch cultures for GUS expression. The yeast colonies were first grown aerobically in YPD medium for 16 hours. The propagated biomass was transferred into expression medium containing promoter activity inducer, galactose, for GUS expression. After 5 hours, yeast biomass was harvested and intracellular protein was extracted. Table 1 illustrates the results of GUS specific activities of ten different transformed clones, indicating that GUS can be highly

Table 1. Transgenic GUS activity in trans- formed starch degrading promoter. The highest specific activity obtained in the culture was 10,057 units per mg of extracted intracellular protein. There was no extracellular GUS activity found since there was no secretion transit peptide sequence to the *gus* gene.

Cone No.	GUS Specific Activity (unit/mg)
Control ^a	16
1	3,469
2	10,057
3	4,061
4	7,013
5	5,309
6	7,786
7	6,035
8	4,439
9	4,000
10	8,346

^aNo genetic transformation

Starch medium was also used to cultivate transformed yeast clones for GUS expression. Corn and waste potato starches were used. The culture medium is a sugar-free medium and contains 1.0% corn or potato starch supplemented with 1.0% peptone and 0.5% yeast extract. After a 2-day growth period in the starch medium, the biomass was collected and GUS was induced in a production medium primarily containing 2% galactose as inducer. Cells were harvested periodically and intracellular protein was extracted. Results indicate that GUS activity reached 13,396 units/mg of extracted intracellular protein after 4-hour induction. The GUS activities leveled off thereafter, indicating stable expression of GUS in the cultures.

E1 endoglucanase expression was conducted by transforming the starch-degrading yeast strain *Saccharomyces* by vector pGA2035. Upon transformation, the transformed yeast colonies were obtained on selective YPD agar media containing antibiotic Zeocin after 3-day incubation at 30°C. Twelve transformed yeast colonies were streak-purified on fresh selective YPD agar plates and single colonies were used in batch cultures for E1 endoglucanase expression. The yeast colonies were first grown aerobically in YPD medium for 16 hours. The propagated biomass was transferred into expression medium containing the promoter activity inducer, galactose, for E1 endoglucanase expression. After a 5-hour inducing period, yeast biomass was harvested and intracellular protein was extracted in the extraction buffer using the glass-bead disintegrating method. Table 2 shows the results of E1 endoglucanase specific activities of twelve different transformed clones. The highest specific activity obtained in the culture was 1,724 units per mg of extracted intracellular protein. However, there was no secreted E1 endoglucanase detected in the culture medium even though a native leader sequence was used in the E1 expression construct. This is probably due to the strong binding activity of the E1 cellulose-binding domain to the cell wall cellulose, inhibiting E1 secretion. Preliminary results showed that the removal of the cellulose-binding domain greatly enhanced E1 enzyme secretion.

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Table 2. Transgenic thermostable E1 endoglucanase activity in transformed starch degrading *Saccharomyces*.

Clone No.	E1 endoglucanase Specific Activity (unit/mg)
Control ^a	7
1	1,724
2	961
3	1,241
4	1,383
5	1,294
6	1,273
7	1,257
8	1,111
9	1,501
10	1,258
11	508
12	1,476

^aNo genetic transformation

ACKNOWLEDGMENT

The authors thank Dr. James R. Mattoon of University of Colorado, Colorado Springs, CO for providing the *Saccharomyces* strain. The authors also thank Dr. Steven R. Thomas of National Renewable Energy Laboratory, Golden, CO for providing the gene of a thermostable cellulolytic enzyme, endoglucanase from *Acidotherrmus cellulolyticus*. This work was supported by the U.S. Department of Energy, Office of Science and Technology, under Contract DE-AC06-76RLO 1830. Pacific Northwest National Laboratory is operated by Battelle for the United States Department of Energy under contract DE-AC06-76RLO 1830.

IN-VITRO ENZYME CATALYZED POLYMER SYNTHESIS: NATURAL CATALYSTS DOING NON-NATURAL THINGS

Richard A. Gross* and David L. Kaplan†

Center for Biocatalysis and Bioprocessing of Macromolecules

*Polytechnic University, Professor and Herman F. Mark Chair, Six Metrotech Center, Brooklyn, NY 11201
(rgross@poly.edu)

†Tufts University, Professor of Chemical Engineering, 4 Colby Street, Medford, MA 02155
(dkaplan1@tufts.edu)

Overview

Our program is focused on the use of enzymes in the synthesis and modification of polymers. Nature provides an extraordinarily diverse range of enzyme catalysts that carry out oxidations, reductions, esterification, amidation, glycolysis, decarboxylation, transamination and many other reactions. Our interests are in determining how to apply natural and engineered enzymes to transformations that result in monomer and polymer synthesis, polymer modification and controlled crosslinking reactions. An extraordinary attribute of enzymes is their ability to provide simple direct routes to products where enantio- or regioselective control is beneficial. Another benefit is the potential of enzyme transformations to provide lower energy pathways to products. Enzyme-catalysis can also circumvent the need for heavy metal catalysts that are environmentally unsafe.

The results of our work on polyester synthesis by lipase catalysis have shown that these methods will be very important in the generation polyesters with functional pendant groups without tedious protection-deprotection steps. *Exploitation of enzyme regioselectivity has also proved important for the preparation of novel block copolymer architectures.* For example, a versatile family of surface active agents have been prepared that consist of a carbohydrate head group attached regiospecifically to either an oligo(ester) or an oligo(carbonate) hydrophobic chain segment. In addition, this structural theme has been used to prepare new multi-arm heteroblock copolymers. Architectural control is an outcome of using natural structures as building blocks in combination with enzyme-catalyzed transformations. Similarly, we have exploited peroxidase and laccase reactions in free radical polymerizations. An outcome of these studies has been the ability to gain control of the structural features of the polymer products through a combination of monomer choice, solvent engineering and enzyme choice. An important component in current projects is the application of accelerated directed evolution along with the appropriate high-throughput assays to develop enzyme-catalysts that have improved catalytic efficiency.

Topics of presentation

1) Propagation kinetics from the ring-opening polymerizations of pentadecalactone (PDL), ϵ -caprolactone (ϵ -CL) and trimethylene carbonate were studied using the lipases from a *Pseudomonas* sp. (immobilized on Celite 521) and *Candida antarctica* (Novozym 435). The experimental results were compared to two living kinetic models describing slow-initiation and slow exchanges between dormant and active species. The use of an interferometric technique is providing new information on the distribution of enzymes and real-time monitoring of polymerizations within an immobilization matrix. Recent work using solvent engineering has resulted in large enhancements in enzyme stability at elevated temperatures and, as a result, rapid polymerization reactions. For example, using recently developed conditions for these polymerizations, it is now possible to reach high monomer conversions within a few minutes

2) Lipase-catalyzed ring-opening polymerization of lactones and carbonates using ethylglucopyranoside (EGP) as a multifunctional initiator were carried out. For example, products of variable oligo(-CL) chain length ($M_n = 450, 2200$ g/mol) were formed by variation of the -CL/EGP ratio. This family of amphiphilic structures was used to prepare heteromulti-arm block copolymers of controlled three dimensional structure.

3) Pendant glycolipids placed along polyacrylate chains are of interest as theological modifiers and as biactive agents that, for example, may function in immunomodulation. A new 6-O-acryl-sophorolipid monomer was prepared by a regioselective lipase-catalyzed acrylation of a sophorolipid macrolactone. The homopolymerization of the 6-O-acryl-sophorolipid derivative and its copolymerization with acrylic acid and acrylamide are reported.

Further exploration of *in-vitro* enzyme-catalysis will undoubtedly lead to a better understanding of the strengths and limitations of this approach. By integration of genetic methods to enhance catalytic activity, we hope to

overcome certain limitations and broaden the impact of these methods.

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CATALYSIS II

RHODIUM CATALYZED HOMOGENEOUS HYDROFORMYLATION IN SUPERCRITICAL CARBON DIOXIDE

D. R. Palo and C. Erkey

Department of Chemical Engineering and Environmental Engineering Program,
University of Connecticut, 191 Auditorium Road, Storrs, CT 06269

ABSTRACT

A novel catalyst, $\text{HRh}(\text{CO})[\text{P}(p\text{-CF}_3\text{C}_6\text{H}_4)_3]_3$, was synthesized for homogeneous catalytic hydroformylation of unsaturated compounds in supercritical carbon dioxide. The incorporation of *para*-trifluoromethyl groups to the conventional hydroformylation catalyst, $\text{HRh}(\text{CO})[\text{P}(\text{C}_6\text{H}_5)_3]_3$, provided enhanced solubility in supercritical carbon dioxide. The catalyst was found to be active for hydroformylation of a wide variety of unsaturated compounds in supercritical carbon dioxide at 50 °C and 273 atm. A rate expression was also developed for hydroformylation of 1-octene at the same conditions.

INTRODUCTION

Over 6 million tons of aldehydes are produced annually by the homogeneous catalytic hydroformylation of olefins¹. The shares of the various aldehydes are: C_3 (2%), C_4 (73%), $\text{C}_5\text{-C}_{12}$ (19%), and $\text{C}_{13}\text{-C}_{18}$ (6%). The catalysts generally employed are of the form $\text{H}_x\text{M}_y(\text{CO})_z\text{L}_n$; the two transition metals utilized are rhodium and cobalt and the most commonly utilized ligands are phosphines (PR_3 where $\text{R} = \text{C}_6\text{H}_5$ or $n\text{-C}_4\text{H}_9$). Production of C_4 aldehydes from hydroformylation of propene is dominated by rhodium based catalysts whereas higher aldehydes are produced mainly by cobalt catalysts. Since rhodium is about 1000 times more active than cobalt, processes based on Rh catalysts operate at significantly lower temperatures and pressures than processes based on Co catalysts. One of the major issues in switching to Rh is the difficulty of the separation of products and catalyst by distillation of the aldehydes. The high boiling points of aldehydes beyond C_7 makes such an operation impractical even under reduced pressure due to thermal stability considerations for the catalyst.

A supercritical fluid (SCF) is a fluid that has been heated and compressed above its critical temperature and pressure. At these conditions, SCFs have densities that are greater than those of gases but comparable to those of liquids, thus enabling them to function as solvents. Using SCFs as solvents may have great advantages in catalyst recovery. The solubility of a compound in SCFs is a strong function of temperature and pressure in the vicinity of the critical point. Therefore, the catalyst, products, and reactants may be separated in an efficient manner through temperature and/or pressure programming. Among the SCFs, supercritical carbon dioxide (scCO_2) is particularly attractive as a solvent since it is non-toxic, environmentally acceptable, inexpensive, readily available in large quantities, and has a low critical temperature and a moderate critical pressure. It is non-flammable unlike some other SCFs such as ethane and propane, thus its use does not constitute a safety hazard. Today, there are many scCO_2 extraction plants operating around the world, indicating the technical and economic feasibility of CO_2 -based processes.

A novel catalyst, $\text{HRh}(\text{CO})[\text{P}(p\text{-CF}_3\text{C}_6\text{H}_4)_3]_3$, was synthesized for homogeneous catalytic hydroformylation of unsaturated compounds in supercritical carbon dioxide. The incorporation of *para*-trifluoromethyl groups to the conventional hydroformylation catalyst, $\text{HRh}(\text{CO})[\text{P}(\text{C}_6\text{H}_5)_3]_3$, provided enhanced solubility in supercritical carbon dioxide. The hydroformylation of a wide variety of unsaturated compounds as well as the kinetics of hydroformylation were investigated at 50°C and 273 atm.

SUMMARY

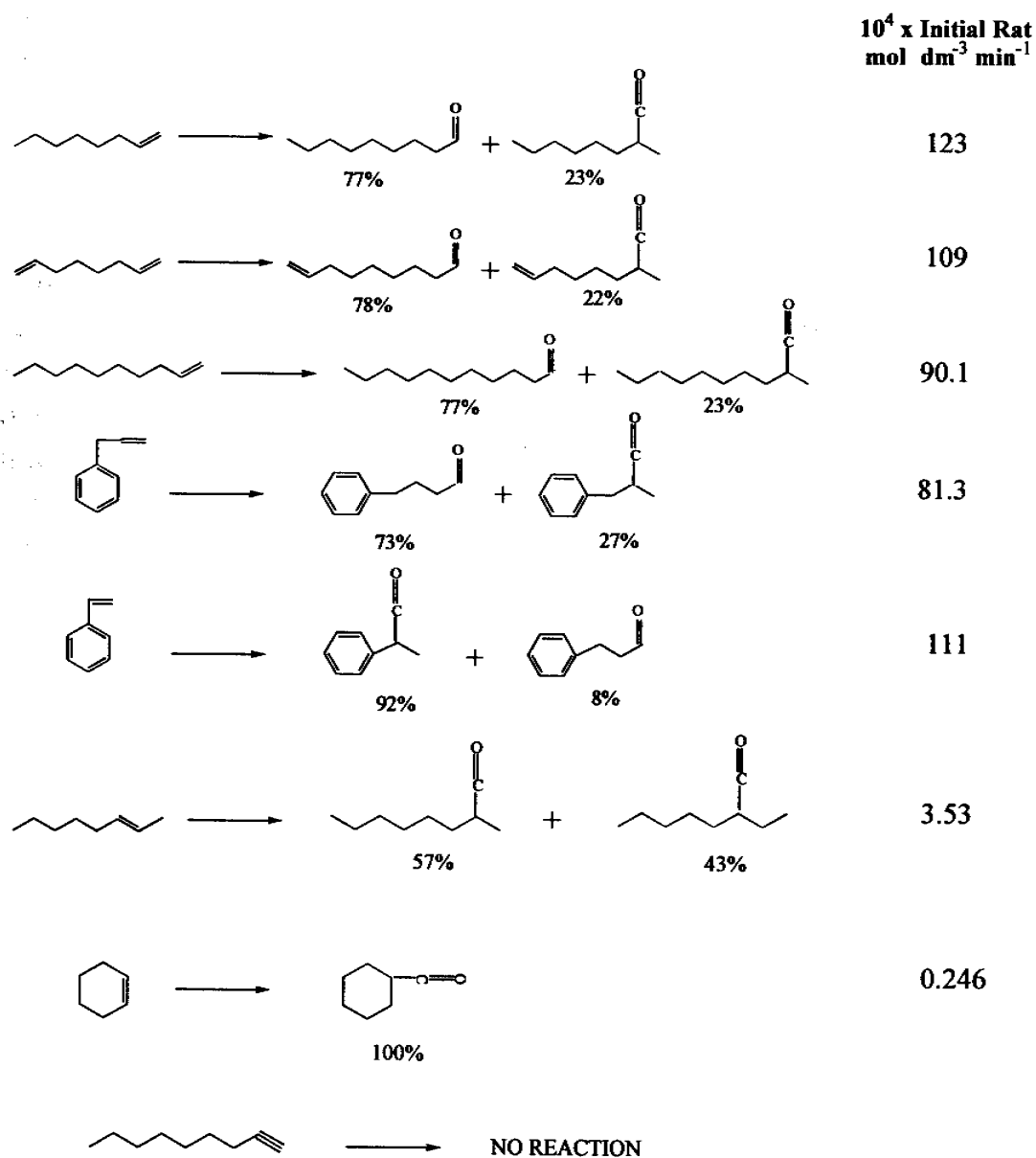
The results are provided below.

The reaction rates for compounds with unsubstituted terminal double bonds were more than an order of magnitude higher than for compounds with substituted or internal double bonds. Furthermore, the reaction rate for cyclohexene was an additional order-of-magnitude lower than for 2-octene, while 1-octyne was not converted at all. The selectivity behavior of the unsubstituted terminal double bonds was similar to that observed previously for 1-octene with *n*:*iso* ratios between 2.7 and 3.5. Hydroformylation of styrene, however, produced an 11:1 ratio in favor of the branched product.

The kinetics of the hydroformylation of 1-octene in scCO_2 was also investigated. The expression which best represented the experimental data was

$$r_{(1\text{-octene})} = \frac{kA^a C^c D^d}{1 + K_B B^b},$$

where $A = [H_2]$, $B = [CO]$, $C = [1]$, and $D = [1\text{-octene}]$, the rate is in units of $\text{mol dm}^{-3} \text{min}^{-1}$, and concentrations are expressed in mol dm^{-3} . The optimized rate parameters were determined to be $k = 6.2 \pm 1.2 \text{ dm}^{2.46} \text{ mol}^{-0.82} \text{ min}^{-1}$, $K_B = 0.69 \pm 0.16 \text{ dm}^{6.6} \text{ mol}^{-2.2}$, $a = 0.48 \pm 0.04$, $b = 2.2 \pm 0.3$, $c = 0.84 \pm 0.03$, $d = 0.50 \pm 0.05$, which yielded a minimized sum of squares value of 3.0×10^{-5} and an average absolute error of 15.5%. The developed kinetic rate expression differs significantly from those previously obtained for the non-fluorinated analogue of the catalyst in organic media. The most significant observations are the ~ 0.5 order rate dependence on $[H_2]$, the lack of substrate inhibition, and the absence of a critical catalyst concentration. This may be due to several factors, including scCO_2 solvent effects, the modified phosphine ligands, and the increased H_2 and CO concentrations relative to conventional systems. These results illustrate the usefulness of scCO_2 as a mechanistic tool for reactions that involve reactant gases such as H_2 and CO , where much higher concentrations can be investigated. Further studies, including *in situ* spectroscopy should shed more light on the observed kinetic behavior. Conditions: $T=50^\circ\text{C}$, $P=273\text{atm}$, $V=54\text{ml}$, $[H_2]_0=[CO]_0=1.1 \text{ M}$, $[\text{catalyst}]=1.2 \text{ mM}$, $[\text{substrate}]_0=0.96$



ACKNOWLEDGMENTS

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EXPLOITING NEAR-CRITICAL REACTION MEDIA IN HETEROGENEOUS CATALYSIS

B. Subramaniam and C. J. Lyon

Department of Chemical and Petroleum Engineering, University of Kansas, Lawrence, KS 66045

This talk will focus on applications of supercritical reaction media in *heterogeneous catalytic systems*. Adsorption/desorption and pore-diffusion are key parameters influencing the activity, effectiveness factors and product selectivity in porous catalysts. Both these parameters can be continuously pressure-tuned in the sc region to obtain unique fluid properties (e.g., gas-like transport properties and liquid-like solvent power), which offer the following advantages: (i) the *in situ* extraction of heavy hydrocarbons (i.e., coke precursors) from the catalyst surface *and* their transport out of the catalyst pores before they are transformed to consolidated coke, thereby extending catalyst lifetime; (ii) enhanced pore-transport of reactants such as hydrogen to the catalyst surface thereby promoting desired reaction pathways; and (iii) enhanced desorption of primary products preventing secondary reactions that adversely affect product selectivity. Complementary experimental and theoretical investigations will be presented to demonstrate one or more of these beneficial effects of pressure-tuning on catalyst activity and product selectivity in a variety of reactions such as hexene isomerization on a Pt/ γ -Al₂O₃ catalyst, Fischer-Tropsch synthesis on supported Fe catalysts and 1-butene/isobutane alkylation on solid acid catalysts. The alkylation application is especially significant since a solid-acid catalyzed alkylation process with enhanced activity offers an environmentally safer alternative to conventional alkylation technology that employs either hydrofluoric or sulfuric acid as catalyst.

HETEROGENEOUS CATALYST DEVELOPMENT FOR HYDROFORMYLATION OF PROPYLENE IN SUPERCRITICAL CARBON DIOXIDE

Srinivas Dharmidhikari, Tim LaPlante, Greg Snyder and Martin A. Abraham

Department of Chemical and Environmental Engineering, The University of Toledo, Toledo, OH 43606

The last decade has heralded a paradigm shift in the way engineers view environmental propriety: waste treatment is no longer an acceptable means of dealing with process wastes. As stated in the Pollution Prevention Act of 1990, the option of first choice is to prevent the formation of waste at the source [Anastas & Warner, 1998]. Clearly, future environmental advances in the chemical process industries will result from re-development of manufacturing processes such that they are environmentally benign.

Many commercially important chemical processes are performed either without a catalyst or with a homogeneous catalyst. If a homogeneous catalyst is used, a significant effort must be made to recover the catalyst so that it can be recycled into the process. The hydroformylation reaction produces greater than 3 billion pounds per year of aldehydes in the United States using a homogeneous rhodium or cobalt catalyst, making this an appropriate target for pollution prevention development.

One of the principles of green chemistry is the elimination of organic solvents and their replacement with benign solvents. Recent experiments have shown that many reactions commercially accomplished with a liquid organic solvent may also be carried out in a supercritical fluid. For example, hydrogenation [Bertucco *et al.*, 1997] and propene hydroformylation [Guo & Akgerman, 1997; Koch & Leitner, 1998] have recently been demonstrated in supercritical CO₂ (scCO₂). Recently, several research groups [Palo & Erkey, 1998, 1999; Koch & Leitner, 1998] have developed prototype homogeneous catalysts that are CO₂-soluble by the addition of perfluorinated chains to the catalytic ligands.

Rather than modifying the catalyst to increase its solubility in CO₂, our research has considered the development

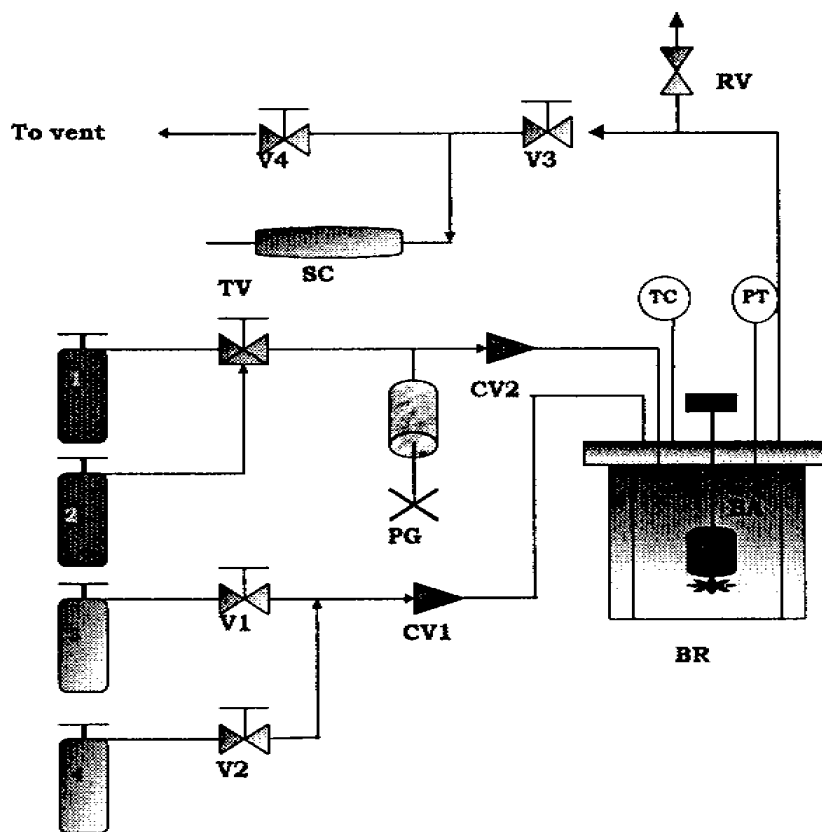
of a heterogeneous hydroformylation catalyst. With the heterogeneous catalyst, recovery is not an issue. Recovery of CO₂ and the aldehyde products can be achieved through a small pressure reduction, allowing for the use of a simple process scheme.

Our development of a heterogeneous catalyst for use with supercritical CO₂ is based on the efforts of previous researchers to develop a heterogeneous hydroformylation catalyst. Numerous supported rhodium species have been shown to be effective for this reaction, but provide varying levels of activity and selectivity. RhCl₃ deposited on silica produces almost exclusively hydrogenation products [Balakos & Chuang, 1995], whereas Rh₄(CO)₁₂ deposited on silica gave 90% selectivity to aldehyde products [Shido, *et al.*, 1995; Ichikawa 1979]. RhCl₃ reduced with NaBH₄ [Lenarda, *et al.*, 1993] and platinum/tin [Homs, *et al.*, 1992] were also reported to provide active and selective catalysts. Rhodium carbonyl complexes supported on SiO₂ [Shido, *et al.*, 1995; Bando, *et al.* 1996] produced by reaction of tris(hydroxymethyl)phosphine with Rh₄(CO)₁₂ in dichloromethane were effective in promoting selectivity to the aldehyde product. High selectivity to the *n*-aldehyde was obtained when a rhodium triphenylphosphine complex was attached to a styrene-divinylbenzene copolymer [Yoneda, *et al.*, 1997]. Rhodium supported on different activated carbons revealed the importance of support and pre-treatment effects in controlling both the activity and selectivity [Kainulainen, *et al.*, 1999].

Experimental

The hydroformylation reaction was carried out in the batch reactor system, shown schematically in Figure 1, and described in more detail in Dharmidikhari [1999]. The system was comprised of the batch reactor, a manual pressure generator (High Pressure Equipment Company) and a sample cell.

Figure 1. Schematic diagram of batch reactor system.

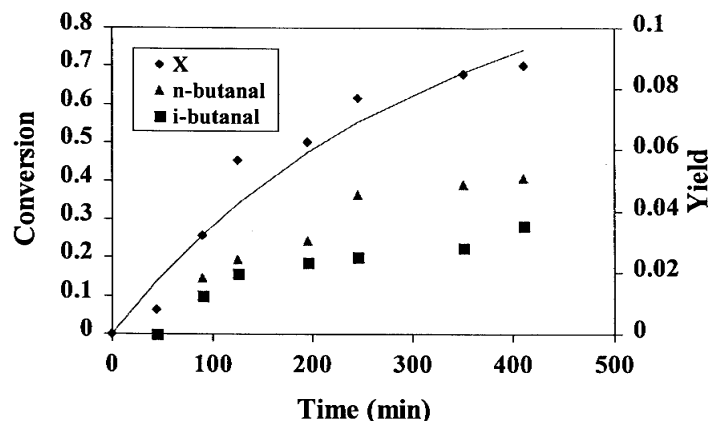


Experiments were performed by loading the desired amount of catalyst in the baskets, purging with nitrogen, and reducing with H₂ at 50 psig and 250°C. Reactants were then added to the vessel based on the pressure required to obtain the desired molar quantities according to the Peng-Robinson equation of state. After the desired operating temperature was attained, additional CO₂ was pumped into the system to give the desired operating pressure (generally ~2500 psia). The analysis of the products was done using an HP 5890 gas chromatograph equipped with a thermal conductivity detector and a 6 ft x.085 in I.D. Porapak T (80/100) packed column.

The catalysts used in this study were prepared using the method of incipient wetness. An aqueous solution of 3 wt % rhodium chloride (RhCl₃•3H₂O, Strem Chemicals) was prepared. Dry activated carbon was weighed and then the aqueous rhodium chloride solution was added dropwise and mixed thoroughly. After impregnation, the sample was dried in air at room temperature. The resultant product was then heated by flowing hydrogen at a temperature of 80°C for 6-8 hours. The catalyst was then reduced by flowing hydrogen for a period of 10-15 hrs at 200°C. After reduction, the sample was allowed to cool and then transferred into a vial and weighed. The difference in weights of the activated carbon before impregnation and after reduction was assumed to represent the amount of rhodium metal on the surface. It was found that the catalyst obtained was approximately 3.2 wt % Rh on Centaur activated carbon and 3.1 wt % of rhodium on PCB activated carbon.

Results and Discussion

Initial experiments using rhodium supported on Centaur activated carbon revealed high consumption of propylene but little formation of product, as shown in Figure 2.



Propylene consumption increased monotonically during the first 350 minutes, after which the consumption appeared to be stable at a conversion of 0.7. The disappearance was modeled assuming first-order kinetics, providing a rate constant of $0.198 \pm 0.025 \text{ hr}^{-1}$; model predictions are indicated by the solid curve in Figure 2. The yield of butanol, defined as the moles of product per initial moles of propylene, increased steadily until the end of the experiment at 420 minutes. The yield of *n*-butanol was always about 50% greater than that of *i*-butanol, with an ultimate yield of approximately 0.06. No other reaction products were observed.

Figure 2. Hydroformylation results using Rhodium on PCB activated carbon at 100°C and 170 bar

Based on the yield of observed products, the total carbon balance was less than 20%, indicating the loss of a substantial quantity of the feed material. However, we were able to recover adsorbed propylene from the surface of the catalyst by reducing with hydrogen at 200°C. Because the experiments were conducted in a batch reactor, these results suggested that propylene could be adsorbed onto the activated carbon support without undergoing any reaction. Confirmation of this hypothesis was obtained by performing an experiment in which the propylene was introduced into the reactor without the addition of CO or H₂. High levels of adsorption were observed, and the adsorption was seen to occur slowly on the support. The rate constant for adsorption onto Centaur activated carbon was calculated to be $0.166 \pm 0.033 \text{ hr}^{-1}$. A second activated carbon (PCB) with larger average pore size was also used in an attempt to reduce the propylene adsorption. The adsorption was decreased, and the first order model provides an estimate of the disappearance rate constant as $0.075 \pm 0.007 \text{ hr}^{-1}$, approximately half that of the Centaur activated carbon.

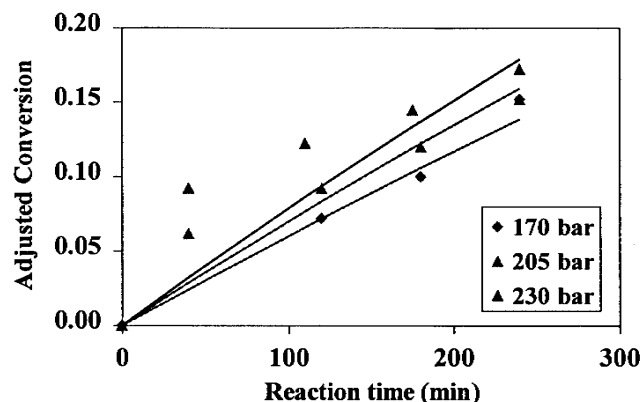
Because of the high level of adsorption, the conversion due to reaction was calculated by subtracting the adsorption, assumed to be equal to that obtained from the first-order adsorption rate constant reported above. With this modification, propylene conversion was approximately 20% for both supports, with roughly equal yields and selectivities for the aldehyde products.

Previous results from homogeneous catalysis suggests that increasing pressure should affect the performance of the catalyst [Guo & Akgerman, 1997]. Experiments with rhodium supported on PCB activated carbon revealed a small effect of total pressure (at constant molar concentration of each of the reactants) on the conversion of propylene, as shown in Figure 3. The change in the values of the rate constants can be modeled using the activation volume, which was calculated as $\Delta v^\ddagger = 142 \text{ cm}^3/\text{mol}$. This value is consistent with activation volumes often reported for reactions in supercritical fluids. Recall that these experiments were performed by increasing the pressure through an increase in the CO₂ loading without changing the propylene loading. Thus, this value of activation volume can be construed as a true measure of the effect of pressure on the rate of propylene adsorption onto the rhodium catalyst supported on PCB activated carbon.

Acknowledgements

This research was supported by the National Science Foundation/Lucent Technologies Foundation Industrial Ecology Research Fellowships program (NSF grant number BES-9873553). Calgon Carbon Corporation provided samples of activated carbon. The support of our research sponsors is greatly appreciated.

Figure 3. The effect of pressure on propylene conversion at 100°C.



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LOW PRESSURE ENVIRONMENTALLY BENIGN ISOCYANATE SYNTHESIS ON Pd CATALYSTS

Steven S.C. Chuang, Pisanu Toochinda and Mahesh V. Konduru

Department of Chemical Engineering, The University of Akron, Akron, OH 44325-3906

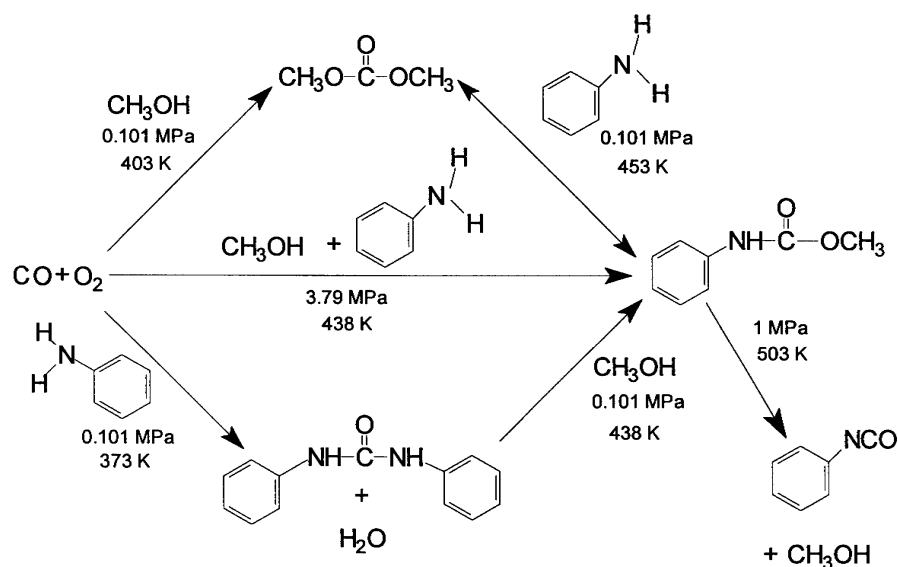
ABSTRACT

Oxidative carbonylation of aniline with methanol to produce carbamate has been recognized as a promising route for isocyanate synthesis. We have recently found that Pd/C with iodide promoter allowed carbonylation to occur at a high rate at atmospheric pressure and 363-438 °K. The use of carbon as a catalyst support for Pd provides a potential heterogeneous pathway for the synthesis reaction. The heterogeneous pathway will eliminate the use of solvent and catalyst recovery step in the synthesis process. This paper reports the mechanistic studies of the carbonylation reaction on carbon supported catalysts and provides the scientific basis for the development of a highly selective and active carbonylation catalyst system.

INTRODUCTION

Phosgene is a very reactive precursor in the synthesis of compounds containing the carbonyl group (-C=O)¹. However, the toxic and corrosive nature of phosgene makes its use a major environment risk. The catalytic carbonylation of aniline/amines and nitrocompounds², the most promising environmentally benign process to replace the conventional method of isocyanate synthesis, provides a direct route to isocyanate without the use of

phosgene. Depending on catalysts and type of reactants used, the process can lead to either isocyanate or carbamate in a single step by either reductive or oxidative carbonylation. Oxidative carbonylation with a



low-energy barrier process and high reaction rate as compared to reductive carbonylation is an attractive alternative for carbamate/isocyanate synthesis. The oxidative carbonylation reaction involving four reactants, CO, O₂, methanol, and aniline, presents the possibility of reaction between CO/O₂, CO/O₂/methanol, CO/methanol, CO/O₂/methanol/aniline, CO/aniline, and others resulting in a complex network of reactions to produce the carbamate species and further the isocyanate species as shown in Figure 1.

Figure 1. Oxidative Carbonylation Network

The formation of carbamate by the reaction of CO and O₂ with aniline and subsequently with methanol (shown in the reaction network in Figure 1) has been reported at 3.79 MPa^{3,4}. We suggest that such high pressures may not be necessary to produce carbamate via this route as it is hypothesized that the reaction of oxygen from methanol and hydrogen from aniline to produce water may provide the necessary driving force for the overall carbonylation reaction to go forward even at atmospheric conditions (0.1 MPa). The objectives of the present study are to investigate the activity of Pd/C catalysts for carbamate synthesis at atmospheric conditions and provide mechanistic information for the complex network of the oxidative carbonylation reaction.

EXPERIMENTAL

5 wt% Pd/C was prepared by incipient wetness impregnation of PdCl₂/HCl (Sigma Chemicals Co.) solution onto activated carbon Amborsorb-563 (Rohm and Haas Co.). The impregnated sample was dried in air at 298 °K for 24 h and calcined at 573 °K for 10 h. The sample was dried in air at 298 °K for 24 h and calcined at 673 °K for 4 h. The ratio of the volume of the salt solution to the weight of support was 1 cm³ to 1 g in the impregnation step during the preparation of both the catalysts. The oxidative carbonylation reaction of aniline was carried out using methanol (CH₃OH), aniline (C₆H₅NH₂), CO, and O₂ with NaI-Pd/C catalyst; the carbamate synthesis from dimethyl carbonate was carried out with dimethyl carbonate, aniline and the catalyst under atmospheric pressure of N₂ over Pd/C. These reactions were studied in a 300-cm³ autoclave (Pressure Products Inc.). The autoclave reactor was pressurized to the desired pressure and subsequently heated to the required temperature.

The reactant/product samples, consisting of a solid and liquid mixture, were sampled periodically from the reactor and bottled. The solid product was allowed to settle and separate from the liquid in the bottle. The solid sample, dried and diluted with KBr (KBr/solid : 9/1), was analyzed by diffuse reflectance infrared Fourier transform spectroscopy (DRIFTS). The liquid was analyzed by transmission infrared (IR) spectroscopy, gas chromatography (GC) with a 6' x 1/8" 12% SE-30 packed column (Supelco Inc.), and a ¹³C nuclear magnetic resonance (NMR) spectrometer.

RESULTS

Figure 1 shows the reaction network and the reaction conditions used for studying each individual reaction. The most interesting of results of these studies are that all the reaction steps can take place over Pd/carbon catalysts with NaI at 0.1 MPa with different temperature requirements.

We have found three reaction pathways leading to methyl-N-phenyl carbamate. The first route involved the reaction of CO/O₂/CH₃OH to produce dimethyl carbonate (CH₃OCOOCH₃) at 403 °K and reaction of dimethyl carbonate with aniline at 453 °K to produce methyl-N-phenyl carbamate. The second pathway is the reaction of CO/O₂/CH₃OH/C₆H₅NH₂ to directly produce methyl-N-phenyl carbamate at 438 °K. The third route is the reaction of CO/O₂/CH₃OH/C₆H₅NH₂ to produce solid diphenyl urea at 373 °K with methanol serving as a solvent.

Increasing temperature from 373 to 438 °K initiated the reaction of the solid diphenyl urea and methanol to produce methyl-N-phenyl carbamate.

The results of this study show the bond breaking process between C of C=O in dimethyl carbonate, diphenyl urea, and methyl-N-phenyl carbamate and the functional group such as phenyl and methoxy is facile, and strongly dependent on reaction temperature.

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Moving Toward Industrial Ecology

June 29 - July 1, 1999

**PROCESS DESIGN
AND MEASUREMENT**

LACTATE ESTERS FOR "GREEN SOLVENTS"—PROCESS TECHNOLOGY ADVANCES

R. Datta, M. Henry, S.P. Tsai, Y. Halpern and J.R. Frank
Chemical and Biological Technology Section, Energy Systems Division,
Argonne National Laboratory, 9700 South Cass Avenue, Argonne, IL 60439-4815

ABSTRACT

Lactate esters produced from carbohydrate have many opportunities and markets as nontoxic replacements for halogenated and toxic solvents. Argonne National Laboratory (ANL) has developed a novel process for the production of high-purity lactate esters from carbohydrates. The process uses advanced electrodialysis and pervaporation technologies to overcome major technical barriers in product separation; more specifically, the process involves cation elimination without the generation of salt waste and efficient esterification that involves reactive separations via membranes. This patented process requires little energy input, is highly efficient and selective, eliminates the large volumes of salt waste produced by conventional processes, and significantly reduces manufacturing costs. The enabling membrane and reactive separation technologies can make it technically and commercially feasible for lactate esters to penetrate these markets for environmentally friendly solvents.

INTRODUCTION

Lactate esters have excellent solvent properties; are nontoxic (approved for food use); are completely degradable; and have the potential to be a major "green" solvent for widespread use in industrial, commercial, and consumer applications. Some published information is available regarding the efficacy of lactate ester solvents and formulations for applications. The potential applications range from major industrial segments (such as semiconductor manufacturing, paints and coatings, metal cleaning, printing, and adhesives) to commercial segments (such as paint stripping and degreasing) and consumer uses (such as general-purpose degreasers and cleaners)^{1,2}. Due to the high cost and limited availability, lactate esters are currently used in specialty applications for precision cleaning, special electronics, and medical applications. Reducing the cost and making the esters available in large volumes could open very large markets for lactate esters, particularly for ethyl lactate.

Ethyl lactate is also a very versatile building-block molecule that can be used to derive a wide variety of chemical products - such as degradable plastic polymers; three-carbon oxygenated chemicals, such as propylene glycol, acrylates, and propylene oxide; and specialty product derivatives - by using certain basic chemical and catalytic conversion processes. The ANL program of chemicals and polymer feedstocks from carbohydrate-derived lactic acid has adopted a platform strategy based on lactate ester as the intermediate, which itself has immediate market potential as a green solvent, and also leads to other products^{3,4}. The potential volume and dollar value of the platform of products that are derivable exceeds \$7-billion lb/yr and \$5 billion/yr in the United States alone^{3,4}. Thus, this platform strategy for these key chemical intermediates can lead to a major opportunity for United States industry to make environmentally friendly chemicals and products from renewable carbohydrate feedstocks.

TECHNOLOGY CHALLENGES

The technical challenges to the production of lactate esters in an efficient, economical, and environmentally friendly process are in the areas of fermentation and product separations and purification. In fermentation, the yield has to be very high, as has to be the product concentration and productivity. Furthermore, the complex nutrient requirements have to be met at a low cost. In some applications, the right product isomer has to be produced⁵.

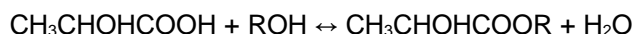
The primary challenges, however, are in the separations and purification. The fermentation process produces a very impure broth of a salt of lactic acid, generally calcium, ammonium, or sodium lactate. However, it is usually the purified acid that is required for conversion into esters or other lactic acid derivatives. The question of how to convert this lactate salt into the acid without consuming an equivalent quantity of another acid and generating an equivalent quantity of waste salt has been the major technical challenge of *cation elimination*. Conventional separation/recovery technologies, such as mineral acid addition or ion-exchange/liquid liquid extraction, produce waste calcium sulfate or other waste salts. For the lactate ester to be suitable for solvent applications or as a feedstock for polymerization or chemical conversion, a very high purity is required. Lactic acid fermentation broth contains high concentrations of impurities; therefore, *product purification* to meet the required specification is a very significant challenge. Although many different approaches have been attempted, esterification has been

known to be the only effective, reliable method to obtain the high purity needed for lactic acid to be a feedstock for the chemical conversion and polymerization processes⁴. However, the conventional processes for lactic acid esterification are inefficient and costly because of the contacting and equilibrium conversion issues.

ANL PROCESS TECHNOLOGY

The ANL process uses advanced electrodialysis and pervaporation for product separation and esterification and overcomes the aforementioned technical barriers. This process requires little energy input, is highly efficient and selective, and eliminates the large volumes of salt waste produced by conventional processes. The ANL process uses a double-electrodialysis approach to recover lactate from the crude fermentation broth and convert the lactate into lactic acid without generating a salt waste. As shown in Fig. 1, the desalting electrodialysis step purifies and concentrates the lactate, and conversion of lactate into lactic acid is accomplished by water-splitting electrodialysis, which employs the bipolar water-splitting membrane. The use of water splitting for fermentation product separation is technically challenging because of the low conductivities of the process streams and the potential interference of soluble impurities to electrodialysis operation. In ANL's double-electrodialysis process, the desalting electrodialysis step improves the operability and efficiency of water-splitting electrodialysis to make such an application feasible and economical. This process was developed and demonstrated with industrial lactate broth. As in the development of many new membrane separation processes, fouling and membrane life were technical issues that had to be addressed. In this technology development a cleaning-in-place (CIP) protocol was shown to control membrane fouling effectively in a long-term test. Furthermore, the stability of membranes over repeated exposures to cleaning solutions was demonstrated.

Esterification is a reversible reaction. The ethyl esterification of lactic acid as shown below has a low equilibrium constant of approximately 2.



Therefore, a high conversion yield cannot be achieved unless the product (ester or water) is removed. However, the relative volatility of the components of this system is as follows:



And so distillation, including reactive distillation, would be energetically inefficient to remove water or ester and achieve a high conversion. In addition, the high volatility of ethanol creates problems in terms of retaining the alcohol in the reaction mixture, and the existence of the alcohol/water azeotrope makes removing the water from the system difficult. As a result, conventional processes for lactic acid esterification with volatile alcohols are inefficient and costly because of intensive energy consumption, low conversion, and high lactate and alcohol losses.

As shown in Fig. 2, the ANL esterification process uses selective pervaporation membranes to efficiently remove water during esterification, whereas alcohol, acid, and ester are retained in the esterification reactor⁶. Commercially available pervaporation membranes have been found to give very good performance for this process, with good membrane stability, economically attractive flux, and a less than 1% loss of alcohol.

This pervaporation-assisted esterification process makes it possible to use a small excess of alcohol to achieve very high conversions and fast reaction rates. Because of the high conversions and low levels of undesirable by-products in the reaction mixture, purification of lactate ester from the reaction mixture becomes very easy. Highly pure ethyl lactate is readily produced at high yields in the subsequent distillations. This lactate ester production process has been successfully demonstrated in the pilot scale, and the ethyl lactate product has been shown to meet the purity requirements of target market applications.

This patented ANL pervaporation-assisted esterification process also allows for a potential breakthrough in the direct esterification of ammonium lactate to ethyl lactate. In this process, the pervaporation membranes pass ammonia and water and yet retain other reactants. By directly esterifying ammonium lactate, this process can potentially eliminate multiple process steps and costs associated with water-splitting electrodialysis or any other acidification process. Pervaporation membranes with higher ammonia fluxes are needed to make this direct process feasible.

The technology has won several awards recently that include the "Presidential Green Chemistry Challenge -

1998" and the Discover Magazine's "Environmental Technology - 1998." NTEC Versol, Inc., of Mt. Prospect, IL has licensed the ANL technology and has entered into a marketing agreement with a major agriprocessing company. Versol™ ethyl lactate is now available in large (tank car) quantities at a reasonable price. Thus, it has become available and affordable for wider applications.

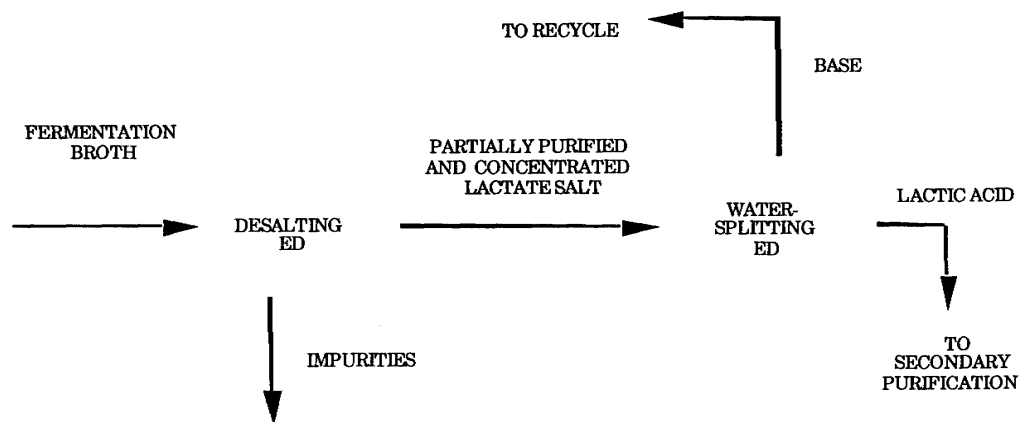


Figure 1. Schematic of the Double Electrolysis Process for Lactic Acid Separation

ACKNOWLEDGEMENT

Work supported by the U.S. Department of Energy, Assistant Secretary for Conservation and Renewable Energy, under Contract W-31-109-Eng-38.

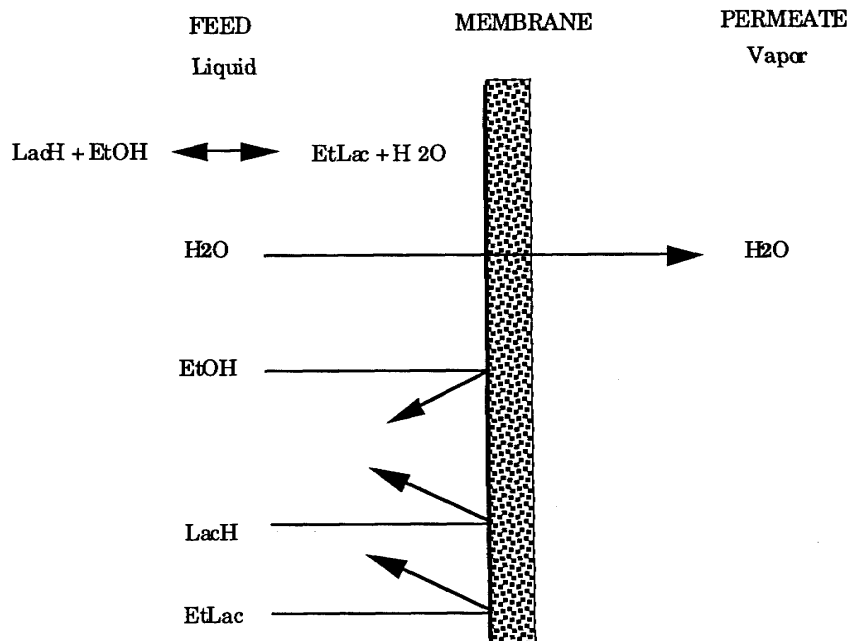


Figure 2. Schematic of the Pervaporation-Assisted Esterification Process

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ENVIRONMENTAL APPLICATIONS OF SUPERCRITICAL FLUIDS AS REACTION MEDIA

J.I. Steinfeld^{*#}, J.W. Tester^{%#}, J.D. Taylor^{%#}, A.G. Casielles^{*#} and P. Bhatia^{*#,1}^{*}Department of Chemistry[%]Department of Chemical Engineering[#]Energy Laboratory

Massachusetts Institute of Technology, Cambridge, MA 02139

¹Present address: Siebel Systems Inc., San Mateo, CA

ABSTRACT

Supercritical fluids have been receiving increased attention as replacements for environmentally hazardous organic solvents. Supercritical water (SCW) is suggested as a medium for waste treatment, remediation, and recycling, and supercritical carbon dioxide (SCCO₂) as a solvent replacement for organic synthesis. In SCCO₂, reaction rates, selectivities, and conversions may be tunable by varying the temperature, pressure, and density of the reaction medium. In order to understand and predict the influence of the supercritical solvent on the rate and selectivity of important synthetic transformations, it is necessary to develop techniques to explore reactions and solvent-solute interactions quantitatively at the molecular level. We will describe progress toward using Raman spectroscopy as a real-time, *in situ* probe for a number of model reaction systems, including Diels-Alder and Kolbe-Schmitt reactions in SCCO₂ and the hydrolysis and oxidation of methyl-*t*-butyl ether in SCW.

INTRODUCTION

Supercritical fluids have been receiving increased attention as replacements for environmentally hazardous organic solvents¹. In order to optimize reaction rates and selectivities of important synthetic transformations in supercritical fluid (SCF) reaction media, it is essential to have a firm understanding of solute-solvent interactions in these high-density fluids². As Seddon has remarked³, in SCFs many of the "old rules of chemistry don't apply, because the thermodynamics and kinetics are different". In addition to providing a convenient real-time, *in-situ* probe of species concentrations in SCFs, optical spectroscopy offers important insights into molecular motions and intermolecular interactions. Representative studies of SCFs using optical spectroscopy include those of Howdle and Bagratashvili⁴, who measured Raman bandwidths of H₂ in supercritical carbon dioxide (SCCO₂) and gave a qualitative explanation of the density dependence; H.L. Strauss and co-workers⁵, who measured Raman bandwidths of H₂ in sub-critical H₂O and carried out a detailed molecular dynamics simulation for this system; Akimoto and Kajimoto⁶, who studied Raman band shifts of acetone and acetonitrile in a variety of supercritical solvents; Kazmian et al.⁷, who studied hydrogen-bonding between (CF₃)₃COH and dimethyl ether in supercritical SF₆ using FTIR; Yamaguchi et al.⁸, who measured spectral shifts of UV/vis absorption and Resonance Raman bands of phenol blue in several SCFs; Xiang and Johnston⁹, who studied the β -naphthol + OH⁻ \rightleftharpoons naphtholate⁻ + H₂O equilibrium in supercritical water (SCW) using UV/vis absorption spectroscopy; and Ryan et al.¹⁰, who studied the β -naphthol + A⁻ \rightleftharpoons naphtholate + HA equilibrium in sub-critical and SCW using steady-state and time-resolved UV/vis fluorescence spectroscopy. Of particular relevance to the study of solvent-solute interactions is the work in Franck's group¹¹ on Raman spectroscopy of HDO in sub- and supercritical water and in MacPhail's group¹² on Raman spectroscopy of cyclohexane-*d*₁₁ in SCCO₂.

Some of the practical advantages of Raman spectroscopy over a technique such as IR absorption include:

- less solvent interference (which in most cases can be subtracted using now-standard spectrum massaging algorithms);
- concentrated solutions can be used without their becoming optically thick;
- IR-inactive modes can be investigated.

On a more fundamental level, Raman band shapes provide detailed information about intermolecular interactions and dynamic behavior¹³. The treatment is essentially based on that given by Gordon in 1965¹⁴, in which it is shown that a Raman band shape is the Fourier Transform of the average motion of the polarizability tensor of the molecule. This approach has been extensively developed in the **W**-matrix formalism for low- to medium-density gases by Millot, Bonamy, and others¹⁵⁻¹⁷, but has not been extensively utilized for Raman band shape analysis in supercritical fluids. The essential data on the time evolution of molecular orientations and polarizabilities can be derived from Molecular-Dynamics (MD) simulations for molecules solvated in SCFs.

EXPERIMENTAL RESULTS

The high-temperature, high-pressure Raman spectroscopic system is based on a Jasco Raman spectrometer,

Model #NR-1000. An argon laser (514.5 nm) is used as the Raman excitation source, and data are collected through a National Instruments Virtual Bench Scope^R interface. The Raman cell is constructed of stainless steel and has an internal volume of 3 ml. Two flat sapphire plates, each 1 mm thick, are used for the optical windows. The temperature of the cell is regulated by a heater-thermostat combination and the pressure is controlled using an expansion and compression pump.

SCCO₂ is an excellent medium for Raman measurements since there are no spectral interferences outside the symmetric-stretch region (1250 - 1400 cm⁻¹). As an example, the totally symmetric ring-breathing (ν_2) and C-H stretching (ν_1) modes of benzene have been recorded at dilutions down to a few mole % in SCCO₂. Distinct band shifts are observed as the solvent reaches critical density.

SUMMARY

Raman spectroscopy is a versatile and powerful technique for probing reactions in supercritical fluids. Work in progress in our laboratory includes determination of reaction rates and elucidation of reaction networks from measurements on reactant and product species concentrations during the course of the reaction, use of MD simulations to model band shapes and band shifts, and extension of the technique to measurements in hydrothermal and SCW reaction systems.

ACKNOWLEDGMENTS

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PROCESS FOR CONVERSION OF CELLULOSIC BIOMASS TO CHEMICALS

Steve Fitzpatrick
Biofine Inc.

The Biofine Technology is based on the thermochemical degradation of cellulose to form levulinic acid. Dilute mineral acid and high temperature are used to produce a rapid conversion of cellulosic feedstock to levulinic acid. The chemistry has been known for around 100 years. However, traditionally, the reaction has given a low yield due to competing side reactions which form waste tars.

Biofine has invented a reactor configuration which favors the desired reaction at the expense of the undesired side reactions. The Biofine reactor system typically produces yields of levulinic acid from cellulose in the range of 60% to 85% of theoretical. Previous methods generated yields in the range of 20% to 40%. Formic acid, and a combustible tar suitable for fuel use, are produced as byproducts.

The Biofine reactor system is a two stage continuous reactor combination. The first stage, a plug flow reactor runs at around 430 Deg. F. and has a residence time of 12 seconds. The second stage, a tank or "backmix" reactor has a residence time of 25 minutes and runs at a temperature of 390 Deg. F. The catalyst is dilute sulfuric acid present in the reaction mixture at around 2% concentration. Biofine has been granted several U.S. and international patents covering this system.

The advantages of this system over previous technology are as follows:

1. Continuous overation - allowing enhancement of the desired reaction to produce levulinic acid, allowing efficient energy integration to reduce energy usage, and maximizing throughput.
2. Short residence time - minimizing the capital cost of the reactor system and allowing high throughput rates with relatively small equipment.
3. Versatility of feedstock - The process can utilize any cellulosic, starchy or sugar feedstock. Feedstocks successfully tested include: waste paper sludge, municipal solid waste, sewage sludge, brewery grains, and agricultural residues such as bagasse, straw, corn stover, cotton waste, etc.
4. Ease of operation - The continuous nature of the process reduces the high labor and maintenance costs associated with periodic batch operation.
5. Plant modularity - The process technology is compact enough to allow large plants (processing 1000 tons per day or more) to be barge-mounted. For plant sites with access to ocean, plants can be constructed on a barge at a central fabrication yard and towed to the site of operation.

Levulinic acid is an extremely versatile intermediate chemical. Up to this time, its high market price (\$5 per pound) has inhibited its large scale use. The commercial potential for this technology is that it can produce a low cost source of levulinic acid which can, in turn, be easily converted to a wide range of useful chemical products with existing or large potential markets. Chemical derivatives include:

- Methyltetrahydrofuran (MTHF), a gasoline substitute or additive and a major component in a U.S. Department of Energy approved alternative gasoline, (MTHF is made by direct hydrogenation of levulinic acid and thus represents the first large scale direct usage of hydrogen as a liquid fuel),
- Delta aminolevulinic acid (DALA), an environmentally benign, highly selective, broad spectrum herbicide,
- Tetrahydrofuran, a high-volume solvent and monomer chemical,
- Diphenolic acid, a substitute for bisphenol A in polymer formulations, and
- Succinic acid, a promising chemical feedstock.

A large scale (two ton per day) continuous demonstration plant has been constructed and operated successfully for over one year. Technical lessons learned from operating experience discussed during the presentation include: control of water balance, appropriate materials of construction, importance of feedstock characterization, and appropriate process control strategies.

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**THE LCAPIX MODULE SOFTWARE:
COMBINING LIFE CYCLE ASSESSMENT WITH ACTIVITY BASED COSTING FOR ENVIRONMENTAL
MANAGEMENT SYSTEMS (ISO 14000) AND CONCURRENT COST REDUCTION**

G.L. Grune, C.O. Grune and S. Sharma
KM Limited, 784 Villier Ct., Virginia Beach, VA 23452

The LCAPIX software module was developed for allowing the user to perform both Activity Based Costing (ABC) and Life Cycle Assessment (LCA). By choosing an ABC approach employing drivers and driver values, the model and relational database provides for a combination of two strategies that enhance the implementation of an Environmental Management Strategy (EMS) consistent with ISO 14000. This approach is necessary for those involved with any aspect of the manufacturing sector - the point source for more than 50% of "undesirable effluents" affecting our global climate.

This software tool has been used to compare different products, processes, and services for their potential environmental burden using different valuation techniques with selected weighted emphasis on issues including, global warming, ozone depletion, acid rain, deforestation, and biodiversity. Case studies showing different techniques used to diminish these burdens while improving internal, external, or "hidden" costs will be described.

3rd Annual Green Chemistry &
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Moving Toward Industrial Ecology

June 29 - July 1, 1999

PHARMACEUTICALS

A NOVEL AND EFFICIENT PROCESS FOR THE PRODUCTION OF CYTOVENE®, A POTENT ANTIVIRAL AGENT

Eric Lodewijk, Ph.D.

President

Yeun-Kwei Han, Ph.D.

Principal Research Chemist

Sam L. Nguyen*, Ph.D., J.D.

Corporate and Patent Counsel

Roche Colorado Corporation, 2075 N. 55th Street, Boulder, CO 80301

George C. Schloemer, Ph.D.

ABSTRACT

Chemists at Roche Colorado Corporation, Boulder Technology Center, located in Boulder, Colorado, developed a novel commercial process for the manufacture of Cytovene®, a potent antiviral agent for the treatment of CMV retinitis and the prevention of CMV disease in transplant recipients. The patented Guanine TriEster Process (GTE), which was registered with the FDA as the current manufacturing process for the world's supply of Cytovene®, demonstrated the following process improvements through technology based designs for pollution prevention: a) Reduction in the number of chemical processing and isolation steps from 6 to 1, b) reduction in the number of reagents and intermediates from 22 to 11, c) demonstrated the recovery and reuse for three out of four major reagents, d) reduced the quantities material usage by 87%, e) doubled the processing throughput of the process, and at the same time, f) increased the overall yield of the product by more than 25%.

INTRODUCTION

The design, development, and implementation of environmentally friendly processes for the large scale production of pharmaceutical products is one of the most challenging aspects of business operations in the pharmaceutical industry. Roche Colorado Corporation (RCC), which operates a manufacturing facility in Boulder, Colorado - a beautiful and environmentally progressive community situated at the foot of the Rocky Mountains - strives to carry out its operations in an effort to ensure complete compliance with federal, state, and local environmental laws and regulations. As part of RCC's goals to establish management and operational systems for the continuous improvement of environmental quality in our business activities, RCC has, in essence, adopted the EPA's basic principles of green chemistry: The development of environmentally friendly processes for the manufacture of pharmaceutical products.

Since the discovery of the potent antiviral agent acyclovir (Zovirax®) by Wellcome in 1974, significant progress has been made toward the development of total syntheses of various purine nucleoside derivatives for the treatment of numerous viral infections. With respect to the purine based nucleosides, a particularly challenging problem that relates to the preparation of these nucleoside derivatives involves the development of efficient reaction conditions for the selective alkylation of guanine. Proposed conditions for the selective alkylation reaction include expensive metal salts such as cesium carbonate and cesium iodide in chlorinated hydrocarbons such as dichloromethane. Alternatively, reaction conditions aimed at increasing the solubility and selectivity of alkylation involving N-2 acylated guanine derivatives have included harsh alkylating conditions such as refluxing dimethylformamide, dimethylsulfoxide, sulfolane, and related high boiling solvent systems.

SUMMARY

In the early 1990s, Roche Colorado Corporation, formerly known as Syntex Chemicals, developed the first commercially viable process for the production of Cytovene® ("The Persilyl Process"). By 1993, chemists at RCC's Boulder Technology Center designed a new and expedient process for the production of Cytovene® (the GTE Process) which, at the time, had an estimated commercial demand of about 50 metric tons per year. Compared to the Persilyl Process, the GTE Process reduced the number of chemical reagents and intermediates from 22 to 11, eliminated the (only) two hazardous solid waste streams, eliminated 11 different chemicals from the hazardous liquid waste streams and, of the 5 ingredients not incorporated into the final product, 4 of the ingredients were efficiently recycled and reused.

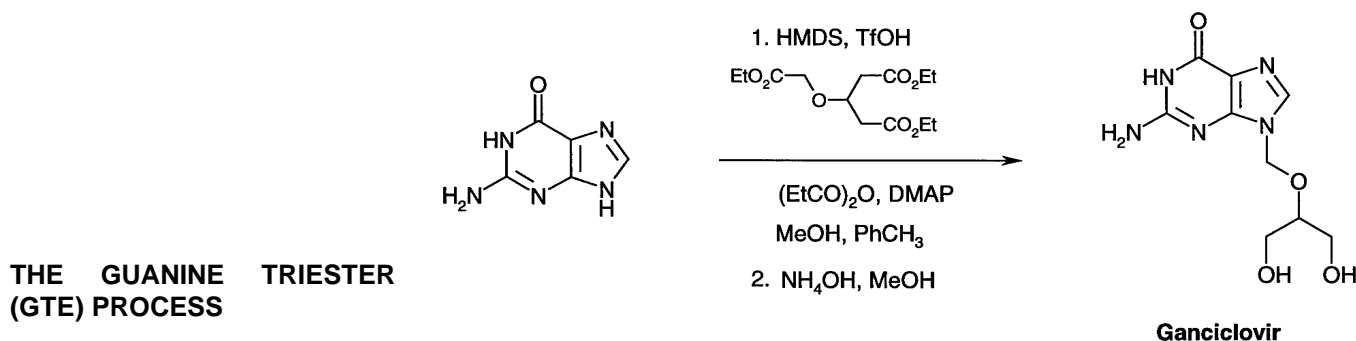
The GTE Process achieved the following process improvements and material savings (at an estimated annual production of 50 metric tons of Cytovene®):

1. Elimination of 3 chemical steps for the preparation of the dibenzyl acetate reagent.

2. Elimination of benzyl chloride (40,000 kg/year), a highly toxic and cancer suspect agent; dimethoxymethane (900,000 kg/year), a flammable liquid and irritant; p-toluenesulfonic acid (2,400 kg/year), a corrosive and toxic reagent; ammonium sulfate (1,500 kg/year); dimethylformamide (3,100 kg/year), an irritant and a cancer suspect agent; dichloromethane (95,000 kg/year), a probable human carcinogen; silica-alumina solid waste (15,000 kg/year); pyridine (3,000 kg/year), a flammable and toxic liquid; xylenes (50,000 kgs/year nonrecycled), an irritant and a flammable liquid; and triethylamine (3,000 kg/year), a flammable and corrosive liquid.
3. Recycle and reuse of ammonium hydroxide (100,000 kg/year).

Significant cost reductions of RCC's business operations realized from the GTE Process provided a positive business case for corporate support of future efforts in designing innovative chemical technologies to reduce the cost for the manufacture of other Roche products worldwide.

The new GTE Process for the commercial production of Cytovene® clearly demonstrated the successful implementation of the general principles of green chemistry: The development of environmentally friendly syntheses, including the development of alternative syntheses utilizing non-hazardous and non-toxic feedstocks, reagents, and solvents, elimination of waste at the source, and elimination of the production of toxic waste and by-products. Furthermore, the process established new and innovative technology for a general and efficient method for the preparation of Cytovene® and other potent antiviral agents for the treatment of CMV retinitis and AIDS related diseases.



REDUCING ENVIRONMENTAL BURDEN THROUGH THE PRACTICAL APPLICATION OF A BIOCATALYST IN PHARMACEUTICAL MANUFACTURING

Benjamin A. Anderson

Lilly Research Laboratories, Chemical Process Development,
 Tippecanoe Laboratories, P.O. Box 685, TL 16, Lafayette, IN 47902

The subject of this presentation is the use of an alternate synthetic pathway designed for the environmentally responsible production of an anticonvulsant drug candidate LY300164. The original synthesis which was employed to support early clinical development proved to be an economically viable manufacturing process. The approach, however, involved several problematic steps. The process required the use of large solvent volumes, chromium oxide (a cancer suspect agent) and led to the generation of disproportional quantities of chromium waste compared to drug produced. These points provided compelling incentive to pursue an alternate synthetic approach.

The new synthetic pathway described herein successfully limited environmental impact by offering a strategy which more appropriately controlled oxidation state adjustments. The new synthesis involved the implementation of several inventive steps on large scale. In particular, keto-reductase activity of a common microorganism was discovered that led to excellent stereocontrol in the asymmetric reduction of a dialkyl ketone. Implementation of the biocatalytic process was enabled on a large scale by employing a novel, yet simple, three-phase reaction system. The protocol overcame long-standing limitations preventing the practical application of yeast-mediated

reductions by allowing high concentrations of the substrate to be charged to the aqueous reaction medium and by providing a facile method for product isolation. An unprecedented autoxidation reaction of a C-1 aryl isochroman which involved the treatment of the substrate with air and sodium hydroxide was also discovered that eliminated the use of transition metal oxidants.

The new process circumvented the use of non-recycled metal and significantly reduced solvent usage. For example, when conducted on a scale to generate 100 kg of LY3000164, the new process avoids the use of approximately 34,000 liters of solvent and eliminates production of approximately 300 kg of chromium waste. Furthermore, the inventive steps of the process represent low cost, easily implemented technology which should find broad manufacturing applications.

SO WHAT DOES GREEN CHEMISTRY/TECHNOLOGY REALLY MEAN TO A PHARMACEUTICAL COMPANY?

David J. C. Constable

Manager, Environmental Product Stewardship

Alan D. Curzons

Manager, Environmental Product Stewardship

Virginia L. Cunningham

Director, Environmental Product Stewardship

SmithKline Beecham, 2200 Renaissance Blvd., Suite 105, King of Prussia, PA 19406

ABSTRACT

This presentation will provide a general overview of several initiatives we are undertaking to determine what Green Chemistry and Technology means for our company. We will describe how we are using Sustainability Metrics (Environmental), an in-depth review of our chemistries, Lifecycle Inventory methods, and total cost assessment (TCA) to answer the very difficult question of what is really "Green." Our findings to date suggest that no single methodology in isolation will yield the "correct" or best result; rather, each different methodology informs the other and provides different viewpoints and potential answers to many difficult questions. A lack of standardized approaches to using these methodologies and differences in impact valuation present very clear challenges in how to best interpret data and make sustainable business decisions. Preliminary results will be presented to show the types of learnings that we are gaining as we have developed our sustainability metrics, pursued a detailed lifecycle inventory of a major drug product, evaluated our chemistries, and collaboratively developed a TCA tool.

INTRODUCTION

Over the last 5 - 10 years there has been considerable discussion about Sustainable Development, Life Cycle Assessment, Total Cost Assessment and Green Chemistry. While many people have been working on various aspects of these initiatives, tools have been developed, and much has been published, there are still considerable hurdles to overcome in operationalizing many of these concepts in an industrial setting. For the past 3 - 4 years, our group has been working to assemble a detailed LCA for a major drug product, develop a practical total cost assessment tool, and establish a viable green chemistry program within SB. Given the particular constraints of the Pharmaceutical approval process, we have been concentrating our efforts on tools, methodologies, and business processes that can be used by scientists and engineers during the R&D process. This has required a considerable up-front effort to reduce the complexity of existing approaches and adapt them to our setting. While this is an on-going process that is still far from completion, we have identified several "quick wins" that will move us closer to achieving sustainable operations while we continue to work on longer term solutions to existing problems.

SUMMARY

This presentation will be composed of three major parts and some concluding remarks. The first part will discuss our work to develop a set of Sustainability Metrics that provide an overall framework for our business process and tool development. We have worked collaboratively with 10 - 15 Fortune 500 companies over the past two years to arrive at a set of core and optional metrics that we are piloting. These metrics cover mass, energy and

pollutants/toxics dispersion, water use and recycling. The complete set will not be reproduced here, but may be found at:

<http://www.aiche.org/cwrt/projects/sustain.htm#Metrics>

The second part will discuss preliminary findings of our product LCI, and how we developed a tool for solvent selection. Our LCI work has been plagued with an almost complete lack of relevant data, and has necessitated a process to develop and validate the data ab initio. The data we have amassed to date suggests that solvents are the largest contributors to life cycle impacts in our manufacturing operations. Appropriate use, reuse and elimination of solvents will profoundly reduce our environmental footprint and safety hazards. Additional insights from the LCI have been gained from the database of chemicals that compose our life cycle supply chain. In the longer term, these insights may affect how we prioritize future E&S efforts, source chemical intermediates, etc.

As a companion effort to our LCI work, we have been working collaboratively with some of the same 10 - 15 Fortune 500 companies as described above over the past several years to develop a Total Cost Assessment methodology and tool. This work is essentially complete, and a manual spreadsheet method will be available in the very near future. This tool complements our LCI work and will allow us to develop cost estimates for impacts not currently easily made visible or quantified.

The third part of this talk will discuss our work on developing tools for green chemistry. We have undertaken a review of our most mature chemical processes that were developed over the past 10 years. The processes have been separated into individual chemical steps and classified into three levels of classification, according to the type of chemistry that is used; e.g., from the general: adding a carbon atom to a ring; to the more specific: a Knoevenagel reaction, a hydrogenation, oxidation, etc. These chemistries are now being evaluated according to a reduced set of metrics we have chosen from the metrics set described previously. This work is allowing us to compare and contrast individual chemistries within and between chemistry types, and provides insights about how these chemistries are combined into chemical processes.

This work is providing a standardized and systematic assessment of the relative "greenness" of our chemistries. Upon completion of our evaluation, we will be able to develop the best tools, based on sustainability principles, that will permit a simple and rapid evaluation of our chemistries and processes. In the medium term we will be able to identify a combination of technologies and chemistries that will drive competitive advantage while ensuring minimal impacts. In the very long term, we can propose areas for academic research where traditional chemistries and chemical process strategies must change if we are to become truly sustainable.

The main conclusion to be drawn from this presentation is that no single tool mentioned above, by itself, is sufficient to answer the many questions concerning how companies are to move towards more sustainable business practices. For our industry, databases containing LCI data do not contain many of the chemicals required to assess the high value-added intermediates used for drug manufacture. TCA is still in its infancy, but offers great promise for a standardized approach to associating economic value with environmental and societal impacts resulting from company operations. Finally, without a standardized and systematic sustainability-driven approach to assessing chemistries and chemical processes, it is, from our perspective, difficult to determine the most sustainable business practices.

3rd Annual Green Chemistry &
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Moving Toward Industrial Ecology

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**GARMENT
LIFE CYCLE**

Extended abstract not received in time for printing.
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**DEVELOPMENT OF RAPIDLY BIODEGRADABLE FABRIC SOFTENER MOLECULES:
THE EFFECT OF ESTER SUBSTITUTION ON THE KINETICS AND MECHANISM OF
BIODEGRADATION OF ALIPHATIC QUATERNARY AMMONIUM COMPOUNDS IN ACTIVATED SLUDGE**

Salvatore T. Giolando, Robert J. Larson and Robert A. Rapaport
The Procter & Gamble Company, 11530 Reed Hartman Highway, Cincinnati, OH 45212

To improve the environmental profile of fabric softener molecules, a program was conducted to develop and commercialize a more rapidly biodegradable version. The process of screening various weak linked aliphatic quaternary ammonium compounds vs success criteria based upon "Practical Biodegradability" is described. Kinetic and mechanistic studies were conducted to characterize the biodegradability of a number of different structural variations around ester substitution of the quaternary nitrogen. Biodegradation was measured using ^{14}C radiolabeled materials at concentrations approximating actual environmental levels.

POLYOXOMETALATE MATERIALS FOR ENVIRONMENTALLY BENIGN CATALYSIS

Huadong Zeng
Graduate Research Assistant
Craig L. Hill
Goodrich C. White Professor of Chemistry
Emory University, Department of Chemistry, 1515 Pierce Drive, Atlanta, GA 30322

ABSTRACT

Monomeric and oligomeric species containing bis(triester) V_6 units, synthesized by reaction of triols with decavanadates in dry polar organic solvents, have been prepared and characterized. Exemplary monomeric complexes, $[(n\text{-C}_4\text{H}_9)_4\text{N}]_2[\text{V}_6\text{O}_{13}\{(\text{OCH}_2)_3\text{CR}\}_2]$, $\text{R} = \text{CH}_3$, CH_2OH , NO_2 , and $\text{NHCO}_2\text{C}_6\text{H}_5$ (structures **1-4**, respectively), are shown to catalyze a reaction of importance in air purification, the aerobic (O_2 -based) oxidation of thiols ($\text{RSH} + 1/2 \text{O}_2 \rightarrow 1/2 \text{RSSR} + 1/2 \text{H}_2\text{O}$) under ambient conditions in acetonitrile solution. Polymeric and other forms of such unusually reactive aerobic oxidation catalysts might form the basis of catalytically air purifying fabrics.

INTRODUCTION

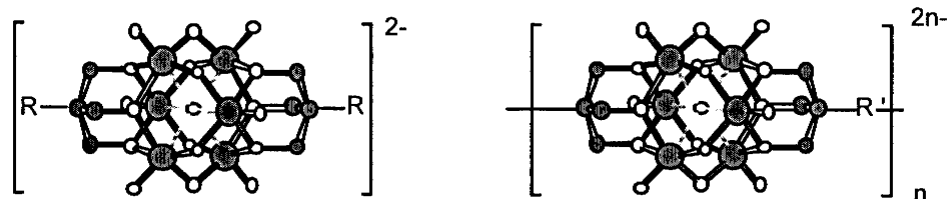
The development of materials such as fabrics or coatings that could catalytically degrade harmful or odorous compounds in indoor air would have an immediate and significant beneficial impact on the environment and human health. Some of the offending gaseous molecules in air include formaldehyde, acetaldehyde, thiols, hydrogen sulfide, ammonia and amines. Formaldehyde, identified as a probable human carcinogen by the International Agency for Research on Cancer and the U.S. EPA, is nearly ubiquitous in our environment (present in building materials, furnishings and other consumer products).¹ Thiols and other sulfur compounds arise from natural gas, pesticide residues and other sources, while ammonia and amines arise from decaying food and other sources. By far the most attractive catalytic materials would be those that use the ambient environment only for destruction of the offending target substances. Clearly the use of liquid-phase decontaminating or disinfecting agents such as hypochlorite (bleach), peroxides, etc. would be neither desirable nor feasible for deployment in durable fabrics and coatings.

Unfortunately the aerobic (O_2 -based) oxidative degradation of the most deleterious volatile compounds is far too slow to be of value. O_2 oxidations generally fall in two regimes under ambient conditions: the ubiquitous reactions involving nearly all organic compounds (plastics, coatings, paints, fats including butter, proteins, etc.), that proceed with half-lives of days to decades or longer, and combustions that proceed violently and nonselectively.

In short, nearly all O_2 reactions are either too slow or too fast; few reactions proceed at controllable intermediate rates of potential value in context with environmental technology.²

We report here the development of catalytically decontaminating molecules and related materials that may offer a route to high-density, high-reactivity catalytically air-purifying fabrics. The molecules and materials of focus are triesters of hexavanadate (henceforth V_6) a representative redox active polyoxometalate (POM) and an oligomer of these tris(ester) V_6 units (Figure 1). POMs are attractive for catalytic and materials applications because their redox potentials, polarities, solubilities, and other properties that bear on such applications can be extensively modified.^{3,4} The only significant developments in chemically reactive fabrics are the recently developed Japanese "self deodorizing" cloths. These materials, sold under trademark names such as "Smoklin" and "Clean Guard", are currently available only in Japan. Thus far, only patents and trade journal reports on these materials are available;⁵ rigorous and informative studies in peer-reviewed journals are lacking. From the literature that is available; however, it is clear that most of these reactive fabrics remove some odor-causing molecules stoichiometrically at best. In other words, they are reagents, not catalysts, and should inactivate quickly and irreversibly. Our laboratory was able to procure one such fabric, "Smoklin", and not surprisingly, given its approximate chemical formula, it was not active. It is noteworthy and ironic that the Japanese industry for these self-deodorizing fabrics has grown to the equivalent of 4 billion US dollars annually despite the reality that many of these materials can't possibly be catalytic and hence can't be of significant lasting value. Importantly, when "Smoklin" as well as other common fabrics were treated with a polyoxometalate (POM) well known to catalyze O_2 -based oxidations under mild conditions, the resulting fabric became quite chemically active.⁶ In short, the demonstration of a material that actually catalyzes fast but controllable oxidations by O_2 and O_2 only under ambient (terrestrial or societal) conditions, would constitute a breakthrough. We report one modest such result here.

Figure 1. Structures of bis-(triester)hexavanadate monomeric (**1-4**; left) and oligomeric (**5**; right) species prepared and characterized. Only the repeating unit in **5** is shown.



RESULTS AND DISCUSSION

Bis(triester)hexavanadate Monomers

Four exemplary complexes of formula $[(n-C_4H_9)_4N]_2[V_6O_{13}\{(OCH_2)_3CR\}_2]$, $R = CH_3$, CH_2OH , NO_2 , and $NHCOC_6H_5$ (**1-4**, respectively; structures shown in Figure 1) were prepared via reaction of tris(hydroxymethylmethane) derivatives with decavanadate in dry polar organic solvents.⁷ Four points suggested that molecules or materials containing these bis(triester) V_6 units might actually function as useful catalysis for aerobic (O_2 -based) oxidations. First, both oxidized and reduced (2- and 6-electron) forms of these units are stable. Second, the hydrolytic stability of the triester linkages to V_3 units (there are two such linkages in the title complexes; see Figure 1) are remarkably stable to hydrolysis (formation of free triol and free polyvanadate does not occur).⁸ Third, a number of organic molecules can reduce the oxidized form of this bis(triester) V_6 species, and fourth, the reduced forms of some bis(triester) V_6 species reoxidize in air to the oxidized forms.

To assess catalytic activity for O_2 oxidations, we chose to investigate the aerobic oxidation of thiols, eq 1, one of the reactions of clear importance in context with developing catalytic air purifying fabrics. Three of the monomeric bis(triester) V_6 complexes were evaluated as catalysts for eq 1, **1**, **2** and **3**. Figure 2 gives the time course of these reactions and the blank (POM-free control reaction) and all reaction conditions. By inference, eq 1 would doubtless also be catalyzed by polymeric forms of bis(triester) V_6 as well, but these experiments have not yet been conducted. The mechanism of these reactions is under current investigation.



Bis(triester)hexavanadate Oligomers

Reaction of the di(trishydroxymethyl) organic linking agent, 1,4- $C_6H_4[CONH(CH_2OH)_3]_2$, with decavanadate in dry *N,N*-dimethylacetamide leads to the oligomer (**5**) illustrated in Figure 1. The isolation, purification and characterization of this complex is given in the Experimental section below. The degree of oligomerization (number of V_6 units per oligomer on average) is 6-7. Future work will seek to increase the reactivity of these systems, establish the mechanism of one exemplary system (probably eq 1 catalyzed by **1**), and explore other

linking diols and triols.

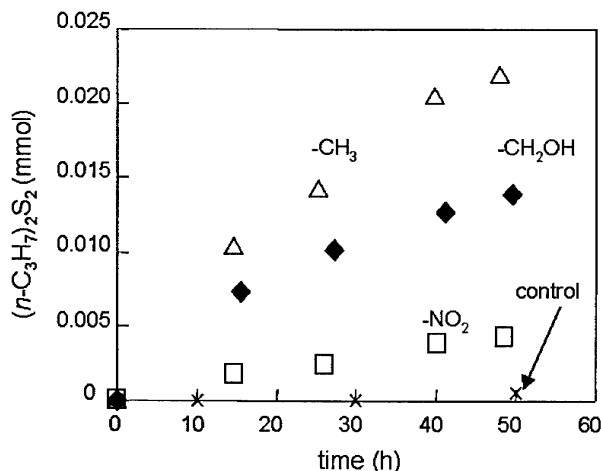


Figure 2. Time dependence of aerobic propanethiol oxidation catalyzed by $[V_6O_{13}\{(OCH_2)_3CR\}_2]^{2-}$, $R = CH_3$ (**1**), CH_2OH (**2**) and NO_2 (**3**) (eq 1). Conditions: 5 mL of CH_3CN solution containing 30 mL of substrate and 5 μL of chlorobenzene (internal standard) were added to 0.0111 mmol catalyst, and the resulting solution was stirred under air at room temperature. The organic compounds were quantified by GC after the reaction times indicated on the figure.

EXPERIMENTAL SECTION

Preparation of $[(n-C_4H_9)_4N]_2[V_6O_{13}\{(OCH_2)_3CCH_3\}_2]$ (**1**)

The compound was prepared using a modification of the literature procedure.⁷ Tris(hydroxymethyl)ethane (0.60 g, 4.0 mmol) was added to a solution of $[(n-C_4H_9)_4N]_3H_3V_{10}O_{28}$ (2.25 g, 1.3 mmol) in CH_3CN (50 mL) with stirring. The solution was refluxed for 5 hr. After cooling, the dark red solution was concentrated to ca. 20 mL via rotary evaporation. A reddish powder (1.8 g) was obtained after addition of 40 mL of ether to the red solution. Recrystallization from CH_3CN /ether (1:1 v/v) gave red crystals in 65% yield. The 1H NMR, ^{51}V NMR and FT-IR indicated **1** was pure.

Preparation of $[(n-C_4H_9)_4N]_2[V_6O_{13}\{(OCH_2)_3CCH_2OH\}_2]$ (**2**), $[(n-C_4H_9)_4N]_2[V_6O_{13}\{(OCH_2)_3CNO_2\}_2]$ (**3**), and $[(n-C_4H_9)_4N]_2[V_6O_{13}\{(OCH_2)_3CNHCOC_6H_5\}_2]$ (**4**)

These compounds were prepared by same synthetic procedure as for **1** using the appropriate tris(hydroxymethyl)methane derivatives and shown to be pure by elemental analysis (for **4**), 1H NMR, ^{51}V NMR and FT-IR.

Preparation of polyhexavanadate oligomer (**5**)

1,4- $C_6H_4[CONH(CH_2OH)_3]_2$ (0.28 g, 0.75 mmol) and $[(n-C_4H_9)_4N]_3H_3V_{10}O_{28}$ (0.84 g, 0.5 mmol) were dissolved in 35 mL *N,N*-dimethylacetamide and refluxed for 24 h. The resulting solution was concentrated to ca. 20 mL at 60 °C. After cooling the solution was added slowly to 80 mL of ether to afford dark red powder (1.0 g). Recrystallization from CH_3CN /ether (1:1 v/v) gave red powder in 50% yield, which was characterized by 1H NMR, ^{51}V NMR and FT-IR. 1H NMR (integration of the coordinated versus the uncoordinated trishydroxymethyl group methylene protons) established that the degree of oligomerization was in the range of 6 to 7.

ACKNOWLEDGMENTS

This work was supported by the U.S. Army Research Office.

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MAXIMIZING THE VALUE OF GREEN CHEMISTRY ACROSS THE GARMENT LIFE CYCLE

Bruce W. Vigon

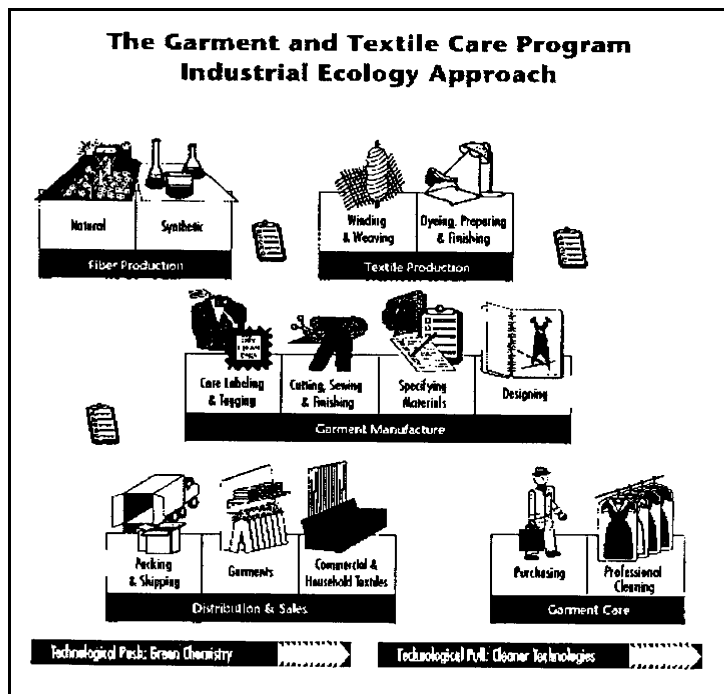
Research Leader

Life Cycle Management Group, Battelle, 505 King Avenue, Columbus, OH 43201

INTRODUCTION

The garment and textile industry consists of a web of producers and users (Figure 1). Using the "value chain" concept helps to reveal the relationships among various groups within the industry. In this concept, investment of labor, technology, and intellectual resources adds economic value to the raw materials and energy inputs. At each life cycle stage, products are realized with greater value to the customer and commanding a higher price when a subsequent market transaction occurs. Decisions made in one part of the system can ripple through the chain and have consequences - some intended, some quite accidental, and some unknown - at many other points. The garment care segment resides at one point in the system, but its activities and their environmental and economic consequences are influenced by activities in many other parts of the web. Conversely, technologies and practices employed by the care segment have the potential to influence technologies, design decisions, and materials selection choices elsewhere in the "upstream" system. Similarly, investments in green chemistry associated with fiber and fabric production can have unintended and unrecognized implications for garment care. The historical approach to understanding these consequences has been incomplete and fragmented, and consequently there has been no way to determine whether the overall environmental and economic situation has improved.

Figure 1. Industrial Ecology Approach



INDUSTRIAL ECOLOGY AS AN ORGANIZING CONCEPT

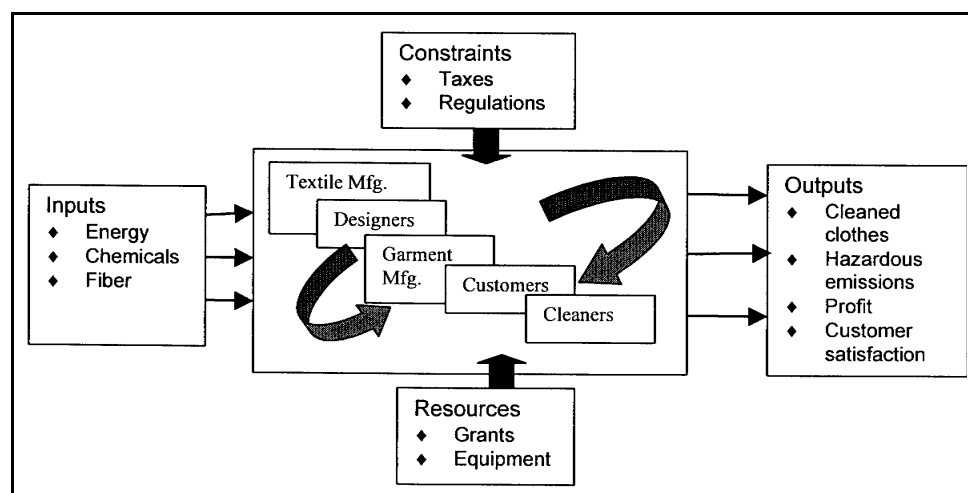
Using an Industrial Ecology (IE) approach, i.e., a systems view that simultaneously evaluates the entire web of industrial activities with respect to economics, performance, and environmental impact, the value contribution of technology insertion at one point can be understood more holistically. This approach enables pollution prevention and increased materials use efficiency through green chemistry initiatives to recognize interrelationships among those groups that design and produce fabrics and garments as well as those groups that purchase, use and clean them.

Environmental effects are a consequence of the flow of material, the use of energy, and the application of technology within the value chain. These activities are influenced by a complex mix of economic forces, business practices, technological options, regulations, scientific understanding, and consumer opinion. The challenge and opportunity of the systems approach is to accomplish the following:

- 1) characterize the potential influences of a change on various points in the value chain,
- 2) identify those changes that promise to enhance the environmental performance of the system as a whole while maintaining its economic viability, and
- 3) provide incentives to encourage infusion of beneficial technologies and/or business practices.

BENEFITS OF A LIFE-CYCLE PERSPECTIVE

- Characterizes the inter-relationships between the garment care segment and the other segments in the value chain (see Figure 2). Helps establish how environmental and financial conditions could be affected by a change initiative, including the insertion of more benign chemistry at one or more points in the garment system.



- Supports efforts to identify issues and trends in different industry segments, and enables the strategies and action plans to be enhanced, grouped and prioritized with regard to their effectiveness and expected impacts.

Figure 2. Industrial Ecology Model Framework

Linking of the environmental and financial "layers" of the IE model allows the impact on both the physical environment (resource demands and burden reductions in amounts of pollution) as well as the financial impacts on profitability to be analyzed in parallel. Because a resource requirement/pollution burden analysis does not capture much of the business value of green chemistry and an economic analysis fails to measure the full environmental benefits, the composite analysis provides the only true measure of the value of such initiatives. Our modeling was performed at a macro level due to the interest in overall national impacts of change initiatives and the simplifying assumptions needed to establish a basis for the economic analysis. Given these interests and limitations, the effects on a single company would not be significant.

THE MODELING METHOD

The particular construct chosen was the Integrated DEFINition (IDEF) modeling method — IDEFO specifically. The IDEFO modeling technique was developed to examine decisions, actions, and activities within a system (Mayer, et al., 1998). The IDEFO description of the garment and textile system was a hierarchical one.

- Level 1 decompositions divide the system into the primary industrial activities
- Level 2 decompositions represent different technology options within Level 1 activities
- Level 3 decompositions model sub-activities within technology options.

As an example, the Level 1 activity "garment care" may be decomposed into two dominant Level 2 cleaning technology categories — wet cleaning and dry cleaning. Level 3 activities associated with wet cleaning include drop off and pick up, pre-treatment, wash and dry, and pressing. Not all activities need to be modeled at the same level of detail. Only those activity nodes where a change initiative is expected to be important need to be decomposed. The effect of activities outside of the garment and textile manufacturing, distribution, and use network, termed "influencing" activities can be incorporated in an IDEFO model. These influencing activities include garment design, research and development, Federal trade policy setting, environmental policy setting, and occupational safety and health policy setting.

Certain assumptions were needed in order to make the exercise tractable. In the physical domain simplifications included:

- Restricting resource inputs to three categories — water, energy, and hazardous materials — to allow a reasonable analysis of materials requirements and regulations for each activity node in the system.
- Restricting outputs of interest to selected products, co-products and hazardous emissions
- Definition of activity constraints that limits or control the relationships between inputs and outputs. These include design specifications or market trends, technology availability, facility capability, and labor skill base.
- Material constraints including physical/chemical properties of materials, occupational and environmental regulations, and waste treatment efficiencies.

In addition to the physical domain the model explicitly shows the financial flows through the system. A simplified set of equations for net profit was used to maximize the use of publicly available information:

REVENUES

Materials - Direct Labor = Gross Profit

Gross Profit - Facilities and Equipment - Administration - R and D = Operating Profit

Operating Profit - Taxes = **Net Profit.**

SUMMARY

A two-stage model development was proposed - an initial descriptive stage and a subsequent quantitative stage.

- Results of the preliminary, descriptive modeling efforts indicated that the decision to focus on key inputs and outputs was a good one. Since the model requires a relationship to be developed for each input-output flow subject to the constraints and available resources, keeping modeled flows to a minimum helps to keep the effort tractable.
- Development of a stakeholder influence concept and a set of influence tables provided a basis for defining decisions/activities to evaluate. The outputs revealed that the model could produce a set of interaction types and a directional set of influence characteristics to provide decision makers with a qualitative indication of how various stakeholders would respond.

The quantitative model has not been completed. This is partly due to the complexity of trying to relate all nodes and flows with a mathematical algorithm and partly due to funding. The descriptive approach with some limited quantitative relationships may be the best balance between investing time and effort to create and calibrate the model and the utility (precision and accuracy) of the results.

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**PESTICIDES AND
AGROCHEMICAL PRODUCTS**

SPINOSAD, A NEW NATURAL PRODUCT FOR INSECT CONTROL

G.D. Thompson

Dow AgroSciences, 9330 Zionsville Rd., Indianapolis, IN 46268

Summary

Spinosad resulted from long standing natural product research programs at Dow Chemical, Eli Lilly and Company and Dow AgroSciences. Spinosad is unique new chemistry produced by fermentation of a naturally occurring soil microorganism. The chemistry met the vision of high levels of efficacy coupled with benign environmental effects but required an extraordinary development and manufacturing effort to meet agricultural volume and cost hurdles. The first product containing spinosad received expedited review by the U.S. EPA and was granted registration as a "reduced risk" insect control product for cotton in early 1997. Spinosad provides highly effective control of many chewing insect pests in numerous crops and ornamentals. Production agriculture is finding it is a highly effective tool for producing more food and fiber on fewer acres and protecting the environment. Spinosad also establishes a new standard for low environmental and human risk and offers new approaches to integrated pest and insect resistance management. It does not leach, bioaccumulate, volatilize or persist in the environment. In hundreds of field trials conducted over several years, spinosad left 70-90% of beneficial insects unharmed. The insect selectivity, coupled with a novel and rapid mode-of-action on many problem insects, allows a scout and treat only as needed approach that results in a powerful IPM tool.

INTRODUCTION

The discovery, characterization and development of spinosad required over twelve years but the vision of an insect control product that was safe, effective, and based on natural products spanned thirty-four years and two companies that formed a joint partnership. Dow Chemical and Eli Lilly and Company merged their plant science divisions in 1989 to form DowElanco. DowElanco evolved into Dow AgroSciences when Dow Chemical assumed full ownership. The soil actinomycete *Saccharopolyspora spinosa* and its metabolite spinosad were discovered prior to the merger at Eli Lilly and Company but there was a parallel natural products effort at Dow. The discovery resulted from a dedicated program with clear product concepts. However, the soil sample that produced *S. spinosa* was collected by a vacationing scientist in the Caribbean, not from professional collections.

The discovery is an amazing story but equally amazing is the development, which required a tremendous effort and commitment to overcome long odds. Hundreds of employees and scores of teams were involved over a twelve year time frame. These included: Discovery Research where entomologists discovered and characterized the initial activity and chemists helped identify the molecule and improve on the activity. Natural Products Research provided the screening samples, isolation and identification efforts and initial production which was a huge effort since titers were initially very low. Technology Development defined the field performance, use patterns, and transferred knowledge to the commercial division. They were also the primary product champions and responsible for justifying continued investment. Formulation Science and Technology developed aqueous based formulations that maintained the environmental benefits and optimized performance. Process Research did not have to work on manufacturing reactions since these were conducted by a microorganism but they had much to do with the manufacturing scale-up and extraction process. The Global Operations division is a collection of smaller departments that detail the important tasks around manufacturing, packaging, supply chain and quality assurance that are required to get the product to market. Global Health, Environmental Safety and Regulatory coordinated the toxicology, environmental fate, risk modeling and regulatory efforts. Commercial received the final handoff and transferred knowledge to the end user and built awareness and value for green properties, but they were also involved early on with product goal inputs. Developing crop protection products is one of the most regulated and complicated product development endeavors. Developing spinosad was even more challenging due to the uniqueness of the chemistry and complexities in manufacturing and would have not been possible without a true team effort focused on a common vision. It is very important that we continue to develop and maintain as many crop protection tools as possible since high production agriculture is extremely important in maintaining the quality of our environment by reducing land requirements for food and fiber production (Avery, 1995).

Spinosad is a mixture of the two most active naturally occurring metabolites (spinosyns A and D) produced by *S. spinosa* (Figure 1, Kirst et al., 1992). Structurally, these compounds are macrolides and contain a unique tetracyclic ring system to which two different sugars are attached. A unique mode of action coupled with a high degree of activity on targeted pests and low toxicity to non-target organisms (including many beneficial arthropods) make spinosad an excellent new tool for management of insect pests.

PHYSICAL PROPERTIES AND ENVIRONMENTAL FATE

Spinosad is a secondary metabolite from the aerobic fermentation of *S. spinosa* on nutrient media. Following fermentation, spinosad is extracted and processed to form a highly concentrated conventional aqueous suspension for ease of use and distribution. Spinosad is a light gray to white crystalline solid with an earthy odor similar to slightly stale water. It has a pH of 7.74, is stable to metal and metal ions for 28 days, and has a shelf life of three years as formulated material. It is considered nonvolatile with vapor pressures around 10^{-10} mm Hg. The degradation of spinosad in the environment occurs through a combination of routes, primarily photodegradation and microbial degradation to its natural components of carbon, hydrogen, oxygen and nitrogen. The half-life of spinosad degraded by soil photolysis is 9-10 days. It is less than 1 day for aqueous photolysis and leaf surface photolysis results in a half-life of 1.6 to 16 days. The half-life of spinosad degraded by aerobic soil metabolism in the absence of light is 9-17 days. Hydrolysis does not contribute significantly to degradation as spinosad is relatively stable in water at a pH of 5-7 and has a half-life of at least 200 days at a pH of 9. The leaching potential of spinosad is very low due to a moderate K_d (5-323), low to moderate water solubility and short residual in the environment. Thus, it does not pose a threat to groundwater (Saunders and Bret, 1997; Anonymous, 1996; Copping, 1998).

NONTARGET TOXICOLOGY

Spinosad is relatively low in toxicity to mammals and birds and is only slightly to moderately toxic to aquatic organisms. In addition, chronic toxicology tests in mammals have shown that spinosad is not carcinogenic, teratogenic, mutagenic or neurotoxic. Spinosad exhibits wide margins of safety to many beneficial insects and related organisms (Schoonover and Larson, 1995). Spinosad has relatively low activity against predaceous beetles, sucking insects, lacewings and mites. It is extremely exciting to have this level of activity coupled with large margins of selectivity for predacious insects an important component of IPM programs. The topical acute activity of spinosad against honeybees is less than 1 µg per bee which places spinosad in the highly toxic to bees category of the EPA. However, once residues have dried completely, toxicity of foraging bees is considered negligible (Mayer and Lunden, 1998).

PHYSIOLOGICAL PROPERTIES AND RESISTANCE MANAGEMENT

Spinosad demonstrates rapid contact and ingestion activity in insects, which is unusual for a biological product. The mode of action of spinosad is characterized by excitation of the insect nervous system, leading to involuntary muscle contractions, prostration with tremors, and paralysis. These effects are consistent with the activation of nicotinic acetylcholine receptors by a mechanism that is clearly novel and unique among known insect control products. Spinosad also has effects on GABA receptor function that may contribute further to its insect activity. This mode of action is unique. Imidacloprid and other nicotinic receptor-based insecticides act at a different site than spinosad. Avermectin, although a natural product and a macrocyclic lactone, also acts at a different site than spinosad. No other class of products affects the insect nervous system with the same mode of action and no cross resistance to spinosad has been demonstrated (Salgado, 1997).

SPECTRUM OF ACTIVITY AND LABELING EFFORTS

Spinosad has been tested extensively on a global basis since 1990 (Carson and Trumble, 1997; Fouche et al., 1998; Kerns, 1996; Linduska et al., 1998; McLeod, 1998; Palumbo, 1997; Riley, 1998; Schuster, 1997; Stansly and Connor, 1998; Walgenbach and Palmer, 1997; Webb, 1998). In general, spinosad provides effective control of pests in the insect orders Lepidoptera, Diptera, and Thysanoptera. It is also effective for some species Coleoptera and Orthoptera that consume large amounts of foliage. Spinosad is generally not effective for control of most sucking insects and mites but some use patterns are being investigated. Spinosad is currently labeled in the U.S. on the brassica vegetable group (broccoli, Chinese broccoli, Brussels sprouts, cabbage, Chinese cabbage - bok choy and napa, cauliflower, cavalo, collards, kale, kohlrabi, mizuna, mustard greens, mustard spinach, Chinese mustard cabbage - gai choy, and rape greens), fruiting vegetable group (eggplant, ground cherry, pepino, pepper, tomatillo, and tomato), leafy vegetable group (including head and leaf lettuce, celery, arugula, chervil, edible chrysanthemum, corn salad, cress, dandelion, dock, endive, fennel, parsley, garden purslane, radicchio, rhubarb, spinach, and Swiss chard), apples, almonds, citrus, potatoes and sweet corn. Crops for which additional U. S. labeling is being pursued include cucurbits, legumes, grapes, stonefruit and numerous minor crops.

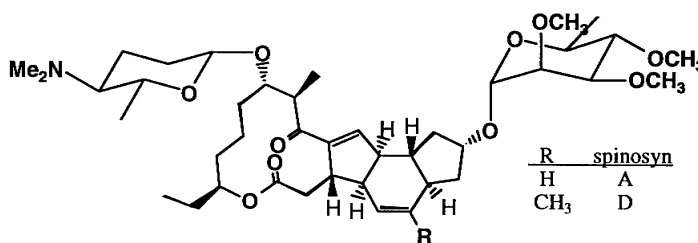
CONCLUSION

Spinosad is a new natural product that not only possesses green chemistry and reduced risk attributes but also delivers very functional insect control that is being rapidly adopted by agriculture. It was the product of a targeted insecticide discovery program that overcame significant hurdles with the dedication of individuals and teams that were united by a common vision to provide improved greener solutions for agriculture.

Figure 1. Spinosad structure.

Molecular Weight: spinosyn A = 731.98,
spinosyn D = 746.00

Empirical Formula: spinosyn A = $C_{41}H_{65}N_{10}$,
spinosyn D = $C_{42}H_{67}NO_{10}$



LITERATURE CITED

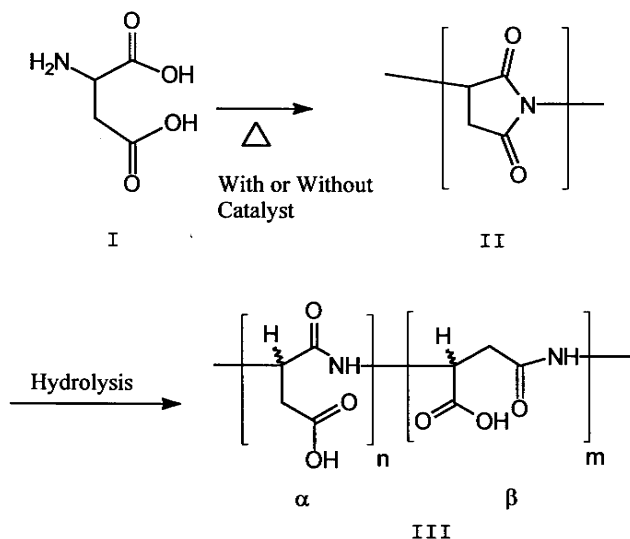
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EFFECTS OF THERMAL POLYASPARTATE ON ENHANCEMENT OF HERBICIDE AND INSECTICIDE ACTIVITY

Ramon Georgis and Robert J. Ross

Donlar Corporation, 6502 South Archer Road, Bedford Park, IL 60501

Thermal Polyaspartate (TPA) is a biodegradable, water-soluble polypeptide obtained via the thermal polymerization of aspartic acid¹. TPA has a variety of uses in water treatment as a dispersant and inhibitor of mineral scale deposition^{2,3,4}, in oil production as a scale and corrosion inhibitor⁵, in personal care products^{6,7}, in agriculture as a nutrient absorption enhancer^{8,9}, and in a variety of pharmaceutical applications^{11,12,13}.



Aspartic acid (I), when heated to a temperature in excess of 180°C, undergoes a solid state condensation polymerization to afford the useful polymeric intermediate known as polysuccinimide (II). Treatment of polysuccinimide with aqueous base, such as sodium hydroxide, affords sodium poly- α,β -D,L-aspartate (III) also known as Thermal Polyaspartate. Acid catalysts, such as phosphoric acid have been added to the aspartic acid to afford higher molecular weight polysuccinimide than is obtained in the non-catalyzed polymerization¹⁴.

Thermal Polyaspartate has been shown to enhance yield and nutrient uptake in agricultural crops when applied with fertilizer. Enhancement of root growth, root hair length and root hair longevity have been reported in many crops by various researchers⁹.

Recent field, greenhouse and laboratory tests have shown that TPA enhances the uptake, and therefore, the efficiency of insecticides and herbicides. (Table 1).

Table 1. Pesticides that have shown enhancement in effectiveness* against selected pests as a result of mixing TPA with their commercial formulations.

Tradename	Chemical Name	Company
Diazinon®	Diazinon	Novartis
Lorsban®	Chlorpyrifos	Dow AgroSciences
Malathion	Malathion	United Agricultural Products
Orthene®	Acephate	Monsanto
Sevin®	Carbaryl	Rhone-Poulenc
Success®	Spinosad	Dow AgroSciences
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Dual®	S-Metolachlor	Novartis
Extrazine®	Atrazine	DuPont
Frontier®	Dimethenamid	BASF
Fusilade®	Fluazitop-N-butyl	Zeneca
Harness®	Acetochlor	Monsanto
Permit®	Methyl-5-3-chloro-1-methyl-1-H-pyrazol e-4-carboxylate	Monsanto
Roundup®	Glyphosate	Monsanto
Touchdown®	Sulfosate	Zeneca

*At label or (and) lower label rates. The first 6 chemicals are insecticides and the last 8 are herbicides.

Thermal Polyaspartate has a high cation exchange capacity (CEC) due to the negative charges associated with carboxyl functional groups of the TPA molecules. The addition of TPA to pesticides applied to soil or foliage, concentrates the molecules of the pesticidal active ingredients, and leads to increase their toxicity to insects and weeds.

In most cases the use of TPA with lower than label rate of pesticides enhanced their speed of action and provided equivalent or better control of the target pest than their label rate. Also in some applications the addition of TPA to the label rate of certain herbicides and insecticides enhanced their speed of action and (or) effectiveness against certain target pests (Tables 2-7). The ability of TPA to attract cations and anions has been reported; it is known that TPA mixed with fertilizers increases their performance by attracting essential nutrient cations such as K^+ , NH_4^+ , Ca^{++} , Zn^{++} , and Fe^{++} . Through a process known as ionic double-layering, nutrient anions such as $H_2PO_4^-$, HPO_4^{--} , NO_3^- , Cl^- , and SO_4^{--} are also attracted to TPA molecules. The high concentration of nutrient cations and anions associated with TPA molecule within the soil solutions enhances the movement of fertilizers (nutrients) to the plant roots through mass flow or ion diffusion.

The use of TPA with fertilizers is exempted from the registration requirements by the U.S. Environmental Protection Agency under provisions of FIFRA. Research showed that ^{14}C -TPA does not enter the plants and therefore its mechanism of activity is outside the plant.

Table 2. Percent mortality of tobacco budworm (*Heliothis virescens*) and cabbage looper (*Trichoplusia ni*) larvae due to foliar application of Success® with or without TPA. 1999 laboratory tests (leaf disks in petri dishes).

Success® + TPA (qt/acre)	Tobacco Budworm		Cabbage Looper	
	24 HAT	43 HAT	17 HAT	41 HAT
0.10 label rate + 0	13.6	47.1	46.2	97.3
0.10 label rate + 1	---	---	90.0*	96.6
0.10 label rate + 2	21.7	86.3*	---	---
0.10 label rate + 4	---	---	72.1*	93.1
0 + 2	1.7	5.6	---	---
0 + 4	---	---	2.5	5.3
untreated check	2.3	9.9	1.9	10.2

*Indicates significant differences between Success® alone and Success® + TPA. HAT = hours after treatment

Table 3. Percent mortality of cabbage looper *Trichoplusia ni* larvae feeding on cabbage due to soil application of Orthene® 75S with or without TPA (2qt/acre). 1999 laboratory studies.

Orthene® + TPA (qt/acre)	18 HAT	44 HAT	68 HAT	98 HAT
0.10 label rate + 0	0.2	50.0	82.6	92.7
0.10 label rate + 2	32.2*	69.4*	93.5*	98.8*
untreated check	0.1	17.2	18.3	38.1

*Indicates significant differences at 0.05 level. HAT = hours after treatment.

Table 4. Percent biomass reduction of giant foxtail (*Bromus rubens*) and velvetleaf (*Abutilon theophrastin*) due to application of four pre-emergence herbicides with or without TPA (2 qt/acre). 1997 greenhouse trials. Urbana, Illinois.

Herbicides (0.25 label rate)	Giant Foxtail		Velvetleaf	
	No TPA	With TPA	No TPA	With TPA
Hardness®	45.0	90.2*	70.7	90.2*
Dual®	61.1	89.8*	---	---
Frontier®	78.3	92.6*	---	---
Extrazine®	62.0	97.7*	29.7	99.4

*Indicates significant differences at 0.05 level, 14 days after treatment.

Table 5. Percent biomass reduction of redroot pigweed (*Amaranthus retroflexus*) and puncture vine (*Tribes terrestris*) due to post-emergence application of Permit® with or without TPA (2 qt/acre). 1999 field trials. Fresno, California.

Treatment	Redroot Pigweed		Puncture Vine	
	3 DAT	21 DAT	3 DAT	21 DAT
Permit® (label rate)				
No TPA	19.6	64.4	30.1	40.3
With TPA	19.1	91.7*	28.7	60.1*
Permit® (0.50 label rate)				
No TPA	19.5	60.0	10.3	36.4
With TPA	18.7	89.7*	28.4	70.0*

*Indicates significant differences at 0.05 level. DAT = days after treatment.

Table 6. Percent biomass reduction of Johnsongrass (*Sorghum halepense*) and Black Nightshade (*Solanum nigrum*) due to post-emergence application of Roundup® Original with or without TPA (2 qt/acre). 1999 field trials. Fresno, California.

Treatment	Johnsongrass		Black Nightshade	
	3 DAT	21 DAT	3 DAT	21 DAT
Roundup® (label rate)				
No TPA	50.1	90.2	20.0	100.0
With TPA	48.9	100.0	46.2*	100.0
Roundup® (0.50 label rate)				
No TPA	30.3	60.7	21.8	69.2
With TPA	48.3	94.0*	58.3*	100.0*

*Indicates significant differences at 0.05 level. DAT = days after treatment.

Table 7. Percent biomass reduction of giant foxtail (*Setaria faberi*) and Johnsongrass (*Sorghum halepense*) due to post-emergence application of Fusilade® with or without TPA (2 qt/acre). 1999 field trials. Fresno, California.

Treatment	Giant Foxtail		Johnsongrass	
	3 DAT	21 DAT	3 DAT	21 DAT
Fusilade® (label rate)				
No TPA	14.1	50.1	25.9	59.8
With TPA	21.7	74.3	19.7	84.3*
Fusilade® (0.50 label rate)				
No TPA	6.0	37.3	13.5	40.3
With TPA	14.7	100.0*	26.1	100.0*

*Indicates significant differences at 0.05 level. DAT = days after treatment.

Thermal Polyaspartate is not expected to be toxic to avian species and honeybees. Because of low toxicity and low exposure, risk to avian endangered and non-endangered species are not of concern. TPA is also considered practically non-toxic to aquatic and terrestrial plants. TPA has low order of acute toxicity and environmental toxicity (Table 8).

Table 8. Summary of TPA toxicological data.

Acute Oral LD ₅₀ (rat)	> 5,000 mg/kg
Acute Inhalation LC ₅₀ (rat)	> 5.1 mg/liter
Eye Irritation (rabbit)	non irritant
Skin Irritation (rabbit)	non irritant
Water Fleas (<i>Daphnia pulex</i>) LC ₅₀	> 1,700 ppm
Fathead Minnow (<i>Pimephales promelas</i>) LC ₅₀	> 2,000 ppm
Algae (<i>Selenastrum capricornutum</i>) LC ₅₀	> 100 PPM

Further development trials are being conducted to determine the cost effectiveness and the practicality of using TPA with pesticides in crop protection management.

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ELECTRONICS

ELIMINATING SOLVENTS FROM SEMICONDUCTOR WAFER MANUFACTURING

Michael Gooch
Conexant Systems, Inc.
Richard L. Bersin
Ulvac Technologies, Inc.

Introduction

In this paper we will discuss the first major utilization of a new process methodology which eliminates the use of solvents in semiconductor 200mm-wafer manufacturing of 0.25u geometry high-speed telecommunications chips. Conexant Systems, a \$1.2 billion company formerly known as Rockwell Semiconductor Systems, has been a manufacturer of these kinds of devices for some years. This application relates to the cleaning of residues left on the wafer following the plasma-etch of metal patterns on a new digital logic chip containing over 1 million transistors. This is a multi-level metal device, requiring several patterning steps utilizing this process.

Conexant Systems attempted to place this product into manufacturing employing conventional hydroxyl-amine based solvents for cleaning. However, this approach resulted in unacceptably low yields of good devices. After trying several other solvent formulations without success, it was decided to investigate this new solvent-free process as a means for elimination of an apparently solvent-based problem while at the same time gaining the benefit of reduced manufacturing costs by elimination of the solvents.

The new process has now successfully been used in manufacturing for over 14 months, 100% solvent free, producing yields of this new device consistently between 96% and 100%. In this paper we will discuss the conventional solvent process; the salient features of the new process; the economic advantages of the new process in reduction of manufacturing costs; the implications for reduced waste disposal burden on the environment; and finally the future benefits to the industry as a whole of adapting this process technology for additional process step as appropriate.

Conventional Solvent-based Processing

The manufacturing process step of interest is used to define the metal lines electrically connecting devices on the chip. The general procedure consists of depositing a metal film on the wafer, typically less than one micrometer thick; coating this film with a photosensitive resist; exposing the wafer to ultraviolet light through a mask, thereby projecting an image into the resist. The resist is then developed, leaving a protective pattern of resist where the metal lines will remain after the etching process.

The wafer is then placed in a plasma etcher, which anisotropically removes the deposited metal from regions not protected by resist. This is done in such a manner that there is essentially no pattern undercut beneath the resist, and the sidewalls of the lines are vertical relative to the wafer substrate surface. This allows for a very high density of lines, capable of handling the signals from greater than one million transistors on the chip.

The plasma etching chamber is filled with chlorine-containing gases flowing at a steady state and maintained at a pressure of the order of a few to a few tens of millitorr. Typical gases are Cl_2 , BCl_3 , N_2 , with total flows the order of 500 to 1000 sccm. A plasma is created within the gas mixture, breaking the molecules down into free radicals and ions. In the plasma the wafer also assumes an electrical potential which is negative in relation to the plasma body (negative bias voltage). This causes energetic ion bombardment of the wafer surface.

These energetic ions bombard both the photoresist that protects the aluminum lines and the open aluminum metal between the lines. The ion bombardment causes erosion and decomposition of the surface of the photoresist (a hydrocarbon material) thereby releasing hydrogen and carbon into the plasma, which have a strong tendency within the plasma to react to form polymers which can deposit on surfaces of the wafer. In addition the ion bombardment of the exposed aluminum surfaces creates compounds of aluminum and chlorine which are volatile (the etching away of the exposed aluminum) and thus are present during the polymer formation. Some of these volatile etch-products become entrained within the polymer during its formation and remain within the polymeric residue on the wafer after the etching is complete.

The ion bombardment of the wafer surface is strongest in the vertical direction since the electric field developed by the voltage bias on the wafer is vertical to the surface of the metal being etched. This energetic bombardment continually removes any polymer which may tend to deposit on the flat exposed metal surface and so the etching

processes continues down through the aluminum film until it is totally removed. However the polymer which forms and deposits on the vertical sidewalls of the lines being etched is not bombarded with such energetic ions and thus remains on the sidewalls of the etched lines.

When the etching process is completed the wafer is then covered with polymer. It is now necessary to remove all traces of this polymeric residue to obtain good electrical performance of the devices and to avoid any corrosion which can occur from reaction of residual chlorine with the aluminum lines.

The conventional process for accomplishing this is to first dry ash the wafer surface, employing a dry Oxygen-based plasma process which burns off the organic materials—typically at temperatures of 250 to 300 degrees C. Although this removes the bulk of residual photoresist and some of the polymeric materials, a substantial quantity of residual non-ashed material comprising metals and complex metal-oxide-organic compounds remains. It is the removal of these last classes of residues which calls upon the use of solvents to accomplish the necessary cleaning.

Common solvents used for this application are N-methyl-pyrrolidone (NMP) containing an amine; an organic-aqueous solution containing ethylene glycol, ammonium fluoride, water, and a surfactant; or a buffered hydroxyl-amine solution containing water and a chelating agent. These solvents are costly to purchase, hazardous to use, and must then be disposed of through conventional hazardous waste-handling facilities. For each 8-inch wafer processed typically 0.05-0.10 gallons of solvent are ultimately consumed.

Typically the wafers are processed in two solvent baths, a 'dirty' bath for initial cleaning and then a 'clean' bath for final cleaning. These are followed by numerous rinses, including finally substantial quantities of DI water for final rinsing to remove all remnants of solvent from the wafer surface.

The New Solventless Process

The new solvent-free process benefits from the fact that most of the metallic compounds incorporated into these polymers are water soluble—e.g. aluminum chloride, TiCl_4 , TiF_4 . This process maintains sufficiently low temperature to avoid oxidation of the metals, yet also can ash organics effectively rendering the entire remaining material soluble in a simple DI water rinse. This is the essence of the new process. This is accomplished through the use of plasma chemical processing with a multiple-step series of processes which are at low temperature and which ash and solubilize residues. The plasma chemical treatment includes steps with RIE plasmas and remote microwave plasma treatments, sometimes in combination, and leaving a dry water-soluble residue.

Applying the New Dry Process at Conexant Systems

A proprietary series of dry processing steps of the type described above are utilized, with the only wet step involved being the final rinse in DI water. Metal level yields between 96% and 100% are consistently obtained. No solvents or other wet chemicals are used in this process and no DI water usage is involved other than the final rinsing when the processing is complete.

Figure 1 shows the electrical resistance of metal lines fabricated with conventional solvent processes compared with the new dry process, with 1.0 representing the normalized results for conventional processing. The new process displays lower mean resistance, plus tighter limits of variation. This reflects the difference between slight corrosion of the metal from the solvents vs. the dry process. Figure 2 illustrates the difference in yield of good devices for the solvent and solventless processes. It is clear that the solvent process yields only about 1/3 the number of good die as the new process.

As new products enter into manufacturing it is anticipated that the percentage of products manufactured employing this process will increase substantially, as the newer smaller and faster products stretch even further the capabilities of solvents to perform the cleaning effectively.

Solvent Usage

Conexant Systems still employs solvent usage for metal cleaning of older products, utilizing equipment which is already in place and proven manufacturing worthy. The hydroxylamine based solvent which they employ is chemically incompatible with other organic chemical agents in use, and thus required the installation of a completely independent waste-handling system for evacuating spent material from the fab and delivering it to the storage tank for waste disposal. This required the installation of a custom \$1.2 million facility for isolating and handling this material.

Spent material is accumulated in a 3,000 gallon tank which is periodically pumped out and delivered by truck to the waste disposal facility for treatment at a cost of \$3.50 per gallon plus \$1,000 for transportation. New solvent is stored in a shed in 55 gallon drums delivered weekly from the supplier. The cost of this solvent new is \$50 per gallon. New solvent is automatically fed from the drums into an active storage container located within the fab building whose level of filling is monitored by constant monitoring of the weight of this storage container which is mounted on a large industrial scale.

Refilling of the tanks in the process equipment in the fabs is controlled and monitored by the computerized system which monitors the conditions of the solvent at critical locations in the delivery system. Again, because of chemical incompatibility the new solvent is stored in the outside shed separated from other chemical storage facilities. Although the quantity of solvent usage by Conexant Systems is proprietary information, experience with other fabs indicates that well over \$1 million is spent annually purchasing these solvents for all reasonably-sized fabs.

Expansion to Via Cleaning

For every metal layer on the wafer there is also a via level above. The Via level is the next most solvent-intensive cleaning process. This is because the etching of the vias with vertical walls, which is essential to sub-micron geometry devices, also involves the creation of polymers within the via on the sidewalls. In the instance of vias the etching chemistry employs fluorine compounds (CHF_3 , C_2F_6 , etc.) to etch the via holes in the silicon dioxide dielectric. The polymers then contain trapped silicon-oxygen compounds from the oxide etching product SiF_4 ; and in addition the polymers may contain small amounts of metal from the bottom of the via, which are backspattered onto the via walls and thereby add metallic contamination to the polymers. Successful processes for solventless via cleaning have already been developed and are in evaluation at Conexant at this time. Expansion of the process technology to via cleaning is presently under consideration.

Overall Semiconductor Industry Benefits

Recent data from Sematech reveals the general level of semiconductor industry indicates over 11 million gallons of solvents are consumed annually by the US industry alone at a cost of over \$½ billion annually on these processes not including the initial cost and amortization of the capital equipment required to carry out the solvent processing.

As the industry converts to these new processes not only will manufacturing costs be reduced, but also a major reduction in the waste disposal burden on the environment will be realized.

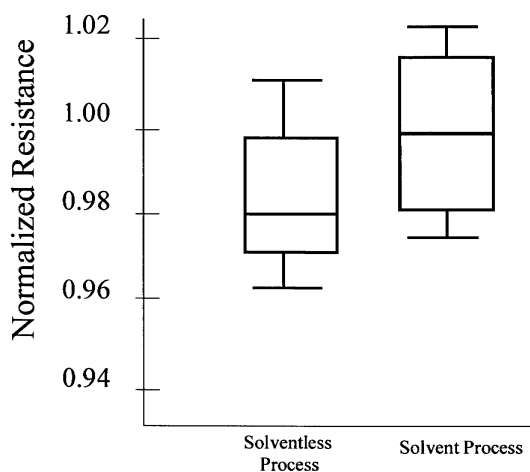


Figure 1. Nominal Line Resistance

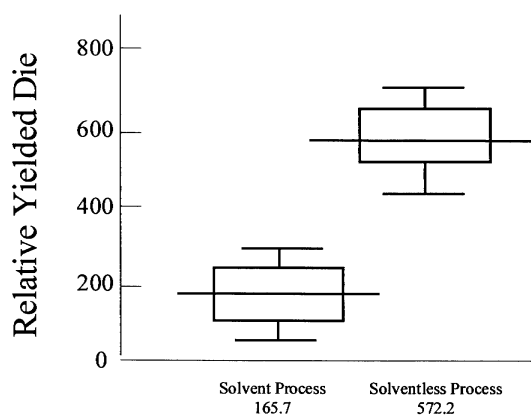


Figure 2. Relative Yielded Die

POLLUTION PREVENTION IN THE MAGNETIC TAPE INDUSTRY: ACRYLATE FORMULATIONS FOR A SOLVENTLESS MAGNETIC TAPE MANUFACTURING PROCESS

David E. Nikles and Jin Young Huh

Center for Materials for Information Technology, The University of Alabama, Tuscaloosa, AL 35487-0209

ABSTRACT

Our objective is to replace the organic solvents used in magnetic tape manufacture with a mixture of liquid acrylates. We have identified commercial acrylates that can serve as the solvent for the coating formulation. Upon electron beam irradiation the acrylates polymerize to give a binder with tensile properties that approach those of commercial solvent-based binders. The solution to a challenging dispersion rheology problem was to treat the magnetic particles with branched silane coupling agents. With a 50 weight percent particle loading the elastic modulus (G') was less than 200 Pa, lower than G' for a conventional, solvent-based formulation. An economic analysis found the operating costs for the solventless process were competitive with the solvent-based process.

INTRODUCTION

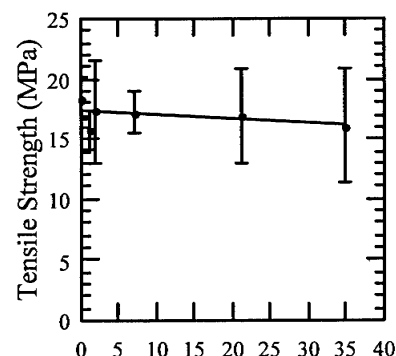
Magnetic tape consists of a magnetic coating on a polyester base. The coating contains magnetic particles held to the base with a polymeric binder and other additives, such as lubricants, carbon black and alumina. Tape is manufactured using a continuous web coating process that uses organic solvents, such as MEK, MIBK, toluene, and THF. MEK, MIBK and toluene are on the EPA's list of hazardous air pollutants. We have estimated that a modern tape coating line uses 600 kg of organic solvents per hour.¹ The atom economy is 30.3 weight percent, with more than 50% of the formulations hazardous air pollutants. The industry meets air emissions regulations by using solvent capture, recovery and recycle systems. Industry sources tell us that 93 to 95% of the solvents are recovered, meaning 5 to 7% escape to the environment. Assuming the coating lines 4000 hr/yr (two eight hour shifts per day, five days per week, 50 weeks per year), then the potential emission would be 120 to 17 metric tons per coating line per year. The objective of our research project is to reduce the air emissions to zero and achieve an atom economy of 100%.

Our approach is to replace the organic solvents with a mixture of liquid acrylate monomers. The monomers would serve as the solvent for the coating fluid. After coating and electron beam irradiation, the monomers would undergo free radical polymerization to form the binder.

ACRYLATE BINDERS

We have identified some promising formulations that contain mixtures containing monoacrylates, diacrylates, triacrylates and acrylate-terminated urethane oligomers.² The aliphatic urethane diacrylate oligomers were Ebacryl 8402, from UCB Radcure, and CN 965 A80, from Sartomer, added strength to the cured coatings. The acrylate monomers were chosen to minimize viscosity, allow fast curing, with low shrinkage and low toxicity. The diacrylates and triacrylates increased the curing speed. The monoacrylates were reactive diluents that lowered the viscosity. The tensile strength for all formulations, except one, exceeded our target of 19 MPa. Only one formation had a Young's modulus that exceeded our specification of 710 MPa. The adhesion problem was solved earlier by irradiating the base film, immediately prior to coating.³ We chose a formulation containing 31% CN 965 A80, 62% propoxylated neopentyl glycol diacrylate (SR 9003), 3.5% highly ethoxylated trimethylolpropane triacrylate (SR 9035) and 3.5% isobornyl acrylate (SR 506A), to begin particle dispersion experiments. A preliminary accelerated aging study of an electron beam-cured acrylate binder formulation, exposed to 60°C and 90% relative humidity, Fig. 1, revealed no significant degradation in tensile strength. We conclude that this class of binders does not present an undue environmental stability problem at this time. This issue will be revisited when the binder materials package is better defined.

Figure 1. Plot of tensile strength as a function of time exposed to 60°C and 90% relative humidity.

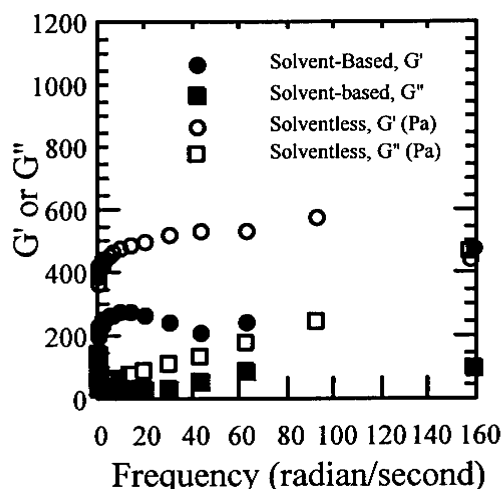


DISPERSION RHEOLOGY

A commercial methacrylate functionalized silane coupling agent from Dow Corning, Z-6030, was covalently bonded to the surface of the iron particles. The original purpose was to use the magnetic particles to

mechanically reinforce the binder by linking the binder to the particles. The commercial iron particles had a ceramic coating containing alumina. We assumed that the surface contains hydroxyl groups that can react with silanol groups from the coupling agent. After treatment the particles had a saturation magnetization of 110 emu/g, a loss of 17%. A TGA curve showed a 12% weight loss upon heating. We attribute the loss in magnetization to the increase in mass due to the surface coating, not a degradation in the particles.

When dispersions were prepared using the particles treated with Z-6030 at 80 weight percent, a thick paste was obtained. The paste had a viscosity of in excess of 10^8 cps, too high for coating, but it did show shear thinning. The viscosity of a typical solvent-based coating fluid is in the range of 10^3 cps, with significant shear thinning. When the surface-treated magnetic particles were used at 50 weight percent, we obtained a coating fluid with



rheological properties, Fig. 2, comparable to a conventional, solvent-based coating fluid. The solventless coating fluid also showed a shear thinning behavior, similar to that observed for conventional coating fluids. The rheological properties of magnetic coating fluids are dominated by the magnetic attraction forces between the particles, which increase the elasticity (G') of the coating fluid. The surface treatment occupied surface sites on the particles that would otherwise adsorb acrylate monomers. The surface coating also provided a steric barrier against particles approaching each other, which decreased the strength of the magnetic attraction between particle, thus decreasing the elasticity of the fluid. This particle surface treatment provides us with a potential means of solving the dispersion and rheology problem.

Figure 2. Frequency dependence of the elastic modulus (G') and viscous modulus (G'') for conventional solvent-based and solventless magnetic dispersions.

ECONOMIC ANALYSIS

We have estimated the costs of our solventless, electron beam cured acrylate magnetic tape manufacturing process. The motivation was to determine whether the operating cost of the solventless process would preclude its adoption by the industry. Earlier we had compared the hourly operating costs of a conventional solvent-based process with our waterborne process and determined that the waterborne process had a 15% lower operating cost.¹ Here we use that analysis as a basis for comparing our solventless process with a conventional process. In Table 1 is a comparison of the hourly operating costs for the conventional solvent-based and our solventless process. In all cases the costs are dominated by the materials costs. For the solvent-based process we considered the case of no recycle and the case where the solvents were captured, purified and recycled. Clearly for the solvent-based process, recycling pays, as it lowers the materials cost to \$1570 per hour while only adding \$13.56 per hour. For the solventless process the hourly materials costs are lower, largely because the solvents are eliminated. By our preliminary analysis the solventless acrylate process is the lowest cost. A problem with this analysis is that we assumed that the cost of the particles was the same for all cases. However, our research has indicated the need to surface treat the particles, which will increase the cost of the solventless process. If the surface treatment increased the particle price by 10%, then the overall cost of the solventless process would be \$1616 per hour, higher than the cost of the solvent-based process with recycle. However, our analysis for the solvent-based process does not include the cost of curing the tape, which is a thermal process done by sitting in a curing room over night. It is difficult to get information about the cost of this process step. The solvents used in conventional magnetic tape coating operations are regulated under the Clean Act Amendments, RCRA and TSCA. In addition to the engineering controls required to safeguard the workers and the environment, these regulations require compliance monitoring and reporting. We assume that an operation that uses our solventless process would be relieved of the reporting requirements which would lower costs. In addition to eliminating the possibility of air pollution, our solventless magnetic tape manufacturing process would also provide better tape at a lower cost.

ACKNOWLEDGMENTS

This project was funded in part by The University of Alabama; Federal funds under the cooperative agreement CR822961-01-0 with the Risk Reduction Engineering Laboratory, U. S. Environmental Protection Agency; Federal funds as part of the program of the Gulf Coast Hazardous Substances Research Center, supported under cooperative agreement R815197 with the EPA, and the U. S. Environmental Protection Agency grant

R826728-01-0. The project used shared instrumentation purchased through the NSF Materials Research Science and Engineering Center award DMR-9400399.

Table 1. Comparison of hourly operating costs.

	Solvent-Based (no recycle)	Solvent-Based (with recycle)	Solventless
Materials	\$2022	\$1570	\$1481
Dryer Energy	\$7.69	\$7.69	\$0
E-Beam Curing	\$0	\$0	\$7.80
Nitrogen	\$0	\$0	\$30
Solvent Recovery	\$0	\$13.56	\$0
Thermal Curing	?	?	\$0
Cost of Compliance	?	?	?
Total	\$2030	\$1591	\$1519

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THE INDUSTRIAL ECOLOGY IMPLICATIONS OF LEAD-FREE SOLDERING

Professor Laura J. Turbini

Materials Science and Engineering, Georgia Institute of Technology, 778 Atlantic Drive, Atlanta, GA 30332-0245

ABSTRACT

A move toward lead-free electronics in Europe and Japan is motivated by the requirements of end-of-life management. When viewed from an industrial ecology perspective, it is important to reflect on lead-free soldering and its environmental impact, which includes processing considerations, energy use, ground water contamination and alloy availability. Based on these criteria, lead-free products are not more environmentally friendly than the present Sn/Pb soldered electronics. Thus, the focus of future regulation should be on recovery of the metals rather than the elimination of lead.

INTRODUCTION

The Surface Mount Council¹ has identified the issue of lead-free electronics as an emerging area for concern and evaluation. This has been triggered by the European Union's (EU) proposal for a *Directive on Waste from Electrical and Electronic Equipment* (WEEE)² and by the Japanese focus on recycling which has resulted in timetables for lead elimination.³ From an Industrial Ecology perspective, it is important to evaluate the environmental impact of the proposed alternatives and to compare those with that of the present Sn/Pb solder.

*Industrial ecology is the means by which humanity can deliberately and rationally approach and maintain a desirable carrying capacity, given continued economic, cultural and technological evolution.*⁴

Based on this definition, it is important to reflect on lead-free soldering from the perspective of its environmental impact which includes processing considerations, energy use, ground water contamination and alloy availability.

LEAD-FREE SOLDERING

A number of Japanese companies are introducing lead-free product to obtain a marketing advantage. Panasonic, for example, is using a proprietary Sn-Ag-Bi-X alloy for their portable mini-discs while Sony has patented a 5-component alloy (Sn-2Ag-4Bi-0.5Cu-0.1Ge) with a melting range of 210-217 °C which they claim provides superior wetting and better mechanical properties.

Nortel has manufactured a totally lead-free Meridian *Desktop Phone* using eutectic Sn-0.7Cu with a melting point of 227°C. The board metallization is copper preserved with an organic solderability preservative (OSP) and component leads are typically Sn-plated or Sn/Cu alloy. The preheat temperatures (~190 - 200°C) and reflow temperatures required (245 - 257°C) are higher than those typically used with Sn/Pb solder (225 - 245°C). Due to moisture in boards and components, coupled with the high soldering temperatures, all boards are pre-baked for 4 hours at 100 - 125°C. Sn/Cu solder has better mechanical properties than Sn/Pb, but there are reliability fall-outs due to board warpage, which becomes more severe when multilayer boards are used. Also, many of the components are not rated for these high temperatures. Automotive manufacturers such as Ford have chosen Sn-3.5Ag eutectic because it enhances reliability for under-the-hood applications.

The UK⁵ has done study of lead-free alloy alternatives and found that the optimum solder replacements are based on Sn-Ag-Cu. Sn-3.8Ag-0.7Cu was recommended for general-purpose use with Sn-3.8Ag-0.7Cu-0.5Sb for wave soldering applications.

PROCESSING ISSUES

The environmental impact of the change to lead-free appears to be significant. Most of the alloys under investigation melt at temperatures that are 30-40°C higher than that of eutectic Sn/Pb solder, although the Sn/Bi and Sn/In alloys melt at much lower temperatures. For the higher melting solders an increased scrap rate is expected due to board warpage. In many cases new substrate materials will be needed due to thermal requirements. Electrolytic capacitors and other components are not rated to experience those high temperatures and as a result failures are anticipated. The specially tailored alloy patented by Sony would present manufacturing problems because it will be difficult to maintain the exact composition in wave soldering.

ENERGY USAGE

Most of the proposed alloys require higher processing temperatures giving rise to a significant increase in energy use for soldering. In addition, the Nortel process requires pre-baking boards and components which greatly increases energy usage.

GROUND WATER CONTAMINATION

One of the main concerns regarding lead-solder has been the leaching of this heavy metal from electronics in landfill. The US EPA has developed a Toxic Characteristic Leaching Procedure (TCLP) to determine if a waste is hazardous. They have developed regulatory limits for many of the elements in these solder alloys (Table 1).⁶

Five lead-free alloys were evaluated according to the leachate test. These include Sn-3.2Ag-0.5Cu, Sn-3.5Ag, Sn-2Ag, Sn-0.7Ag and Sn-5Sb. The findings have important implications for environmental evaluation of these alloys:

- Sn-5Sb leachate contained 10,000 times the allowable limit of Sb;
- SnAgSb alloys leach above the allowed limit;
- SnAg alloys are close to the EPA limit;
- Sn did not leach significantly because most tin salts are not soluble in water at room temperature;
- Lead leachate from circuit boards soldering with Sn/Pb solder contained less than 40 times the allowable level of Pb.⁷

Table 1. Regulatory Limits

Element	Media	Limit, mg/L	Source
Tin	All	None found	
Silver	Drinking Water	0.10	EPA 40 CFR 141
Silver	TCLP Leachate	5.0	EPA 40 CFR 261
Antimony	Drinking Water	0.006	EPA 40 CFR 141
Antimony	TCLP Leachate	1.0	TRCC 30 RAC 355*
Copper	Drinking Water	1.0	EPA 40 CFR 141
Copper	TCLP Leachate	500	Various State Regulations
Lead	Drinking Water	0.015	EPA 40 CFR 141
Lead	TCLP Leachate	5.0	EPA 40 CFR 261**

*Texas statutes

**Some jurisdictions observe a 1.5 mg/L limit based on a multiple of the 0.015 mg/L drinking water limit.

ALLOY AVAILABILITY

The issue of alloy availability is an important one. Many of the proposed substitutes such as bismuth, silver and indium are mined with lead ores and their world reserves are limited. Comparing the base reserve of lead - 130 million metric tons and copper - 590 million metric tons against antimony - 4,700,000 metric tons, silver - 420,000 metric tons, bismuth - 250,000 metric tons, and indium - 5700 metric tons makes it obvious that the supply of lead-free alloy are limited.⁸

SUMMARY

Lead-free soldering has been studied for several years in hope of finding a drop-in replacement for Sn/Pb solder. None have been found. Those alloys presently used in or proposed for lead-free electronics have a number of problems from an industrial ecology perspective. Most of them require higher processing temperatures, which may lead to board warpage, component failures and higher reject rates. Energy usage for manufacturing increases substantially, and there is evidence that metal leachate from alternate alloys in many cases exceeds the regulatory limit. Finally, many of the proposed alternatives are resource limited and could not keep up with demand.

An alternate approach to eliminating the potential hazards of lead is to execute methods for metal recovery and reuse. Our natural resources are dwindling and we have a responsibility to preserve them for future generations.

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POLYMERS

WATER BASED LIQUID DISPERSION POLYMERS

M. Ramesh

Technical Director, Global Polymer Science

P. G. Murray

Group Leader, Global Polymer Science

Nalco Chemical Company, One Nalco Center, Naperville, IL 60563

Abstract

High molecular weight water soluble polymers derived from acrylamide are widely used as process aids and water treatment agents in a variety of industrial operations. Historically, these polymers have been produced in liquid form as water-in-oil emulsions. These water-in-oil emulsions, or "latex" polymers, require that large amounts of oil and surfactants be employed in their manufacture. These oils and surfactants allow the polymers to be manufactured as liquids, but they play no role in the end use application. As a consequence, they are eventually introduced into the environment along with the active polymer component. In order to overcome the disadvantages associated with the production of these polymers as water-in-oil emulsions, Nalco has developed and commercialized a unique homogeneous dispersion polymerization technology to manufacture these important polymers as liquids in aqueous salt solution. This technology provides for a number of environmental advantages associated with both the manufacture and the application of these polymers.

Introduction

Water is a primary raw material or processing aid in most industrial operations, including paper manufacturing, oil production, municipal water clarification and waste treatment, mining and mineral processing, petroleum refining, metalworking, steam generation, and food and beverage production. Because of its unique physical properties and relative abundance, water is used in all of these industries to transport solids economically or to transfer heat efficiently. Typically, water taken from the environment for use in these operations needs to be conditioned to remove suspended solids, organic matter and other materials that may be detrimental to the process. Likewise, water that is being used in these industrial processes must often be treated at some point to effect a solid/liquid or a liquid/liquid separation, which is often critical to the efficiency of the central operation. Finally, water that has been so used must often once again be treated prior to its return to the environment in order to remove harmful wastes and contaminants (oils, sludges, metals, etc.). Synthetic organic polymers are widely used for all of the above listed purposes. The majority of these polymers, which are now consumed at the rate of several hundred million pounds per year, with a market value of approximately three billion dollars, are produced as high molecular weight copolymers of acrylamide.^{1,2}

Until recently, the manufacture of high molecular weight acrylamide based polymers or copolymers in liquid form was accomplished by making water-in-oil emulsions (or "latex" polymers), consisting of a hydrocarbon solvent for the continuous phase and various surfactants to provide emulsion stability. Polymers prepared in this fashion are generally 20% to 40% active polymer. As liquid products, the water-in-oil emulsions are easy to handle, and, *given the proper equipment*, relatively easy to prepare for their end use. Despite these advantages, the water-in-oil emulsions do have several inherent disadvantages. For example, the hydrocarbon continuous phase and surfactant systems that enable their manufacture as liquids play no role in the end use application of the active polymer. As a consequence, at the current consumption levels of about 80 million pounds of active polymer per year, about 90 million pounds of these oils and surfactants are introduced into the environment along with the active polymer, since, until now, no improvements in the manufacture of these polymers as liquids have been forthcoming.³ Moreover, at the end use application, these water-in-oil emulsions are inverted, or diluted with water, and the emulsion is destabilized by the addition of yet another surfactant in order to prepare the polymer component for use. This step requires fairly specialized and relatively expensive high-shear mixing equipment.

Therefore, in order to overcome the difficulties associated with inverse emulsion water treatment polymers, Nalco has developed a unique technology for the manufacture of the same high molecular weight, water soluble polymers in liquid form. This novel polymerization process permits the manufacture of these extremely useful polymers as water continuous dispersions. Thus, the polymer products are still liquids, but they are manufactured in aqueous ammonium sulfate solution instead of in a hydrocarbon and surfactant-based matrix. This technology, and the attending advantages, will be described herein.

Summary

Nalco's new water continuous polymer products are produced through a unique homogeneous dispersion polymerization. In this process, the water soluble monomers are dissolved in aqueous solutions of ammonium sulfate. This solution is then polymerized using a water soluble free radical initiator. During the initial phases of the polymerization, the aqueous salt solution becomes saturated with growing polymer chains, so that when some critical concentration is reached, phase separation occurs and the polymer precipitates into fine particles saturated with monomer and initiator. Further polymerization continues until the available monomer is exhausted. These particles are prevented from agglomerating by the addition of a relatively low molecular weight stabilizer (dispersant) polymer, the structure and concentration of which are critical to the success of the process.

At the end use application, this dispersion is added to water, which dilutes the salt and allows the polymer particle to dissolve rapidly to form a clear, homogeneous solution. The polymers which can be made using the water continuous technology include cationic copolymers of acrylamide (with quaternary salts of acrylate esters or diallyldimethylammonium chloride), anionic copolymers of acrylamide (with acrylic acid or 2-acrylamido-2-methylpropanesulfonic acid) or non-ionic polymers (with acrylamide or vinylformamide).^{4,5} Indeed, almost every water-in-oil emulsion polymer presently available from water treatment chemical companies worldwide can be made in water continuous dispersion form, which demonstrates the broad applicability of the technique.

The environmental benefits associated with the manufacture and application of these water-based dispersion polymers are summarized below:

Elimination of the hydrocarbon solvent and surfactant "carrier" of water-in-oil emulsion polymers used for water treatment

Two cationic copolymers of acrylamide are widely used for a variety of industrial water clarification and separation operations ranging from papermaking to mining to municipal sludge conditioning. The first polymer, Polymer A, is a 10 mole % dimethylaminoethyl acrylate methyl chloride quaternary salt (DMAEA.MCQ)/90 mole % acrylamide copolymer, and the second polymer (Polymer B) is a 30 mole % diallyldimethylammonium chloride/70 mole % acrylamide copolymer. *By choosing to manufacture both Polymer A and Polymer B in water-based dispersion form, instead of as water-in-oil emulsions, over one million pounds of hydrocarbon solvent and surfactants have been conserved by Nalco since 1997 on these two products alone.*⁶ This is over one million pounds of oil and surfactant that would have otherwise eventually been introduced into the environment as a result of various water treatment operations throughout the country. These two Nalco polymer products are manufactured and used in Europe as well as in the United States, resulting in a worldwide impact of the new technology upon the global environment.

The application of an industrial process by-product (ammonium sulfate) as a raw material in water-based dispersion polymerization

As an examination of the patents which have been issued to Nalco will indicate, a variety of inorganic salts will function to cause the formation of stable colloidal particles of polymer in water.⁷⁻¹⁰ However, ammonium sulfate is somewhat unique among the inorganic salts, since it is primarily derived as a by-product from the production of caprolactam, the precursor to nylon. In fact, the conventional caprolactam processes produce anywhere from 2.5 to 4.5 tons of ammonium sulfate for every ton of caprolactam!¹¹ Ammonium sulfate has been widely used as a fertilizer, this providing an effective method for "disposing" of the ammonium sulfate produced by the caprolactam processes. However, as more concentrated nitrogen fertilizers were developed, ammonium sulfate now represents only 6% of the nitrogen based fertilizer consumption.¹² As a consequence of this, and since little ammonium sulfate is used industrially, this salt becomes attractive as a raw material for the production of water-based dispersion polymers. *For example, in 1998 over 3.2 million pounds of caprolactam produced ammonium sulfate was used by Nalco manufacturing and recycled into high performance, environmentally responsible water treatment polymers.*

Wastewater treatment options for operations which cannot afford large capital investments in polymer feeding equipment

One of the chief attributes of the water-based dispersion polymers is that they dissolve readily in water. No "inverter" surfactants or high-shear mixing equipment is necessary, as with the inverse emulsion polymers. While this benefit is universally recognized as a technological improvement, it is particularly advantageous for smaller and medium-sized operations which have not been able to treat their wastewater streams owing to the relatively high cost of the emulsion polymer "inversion" and feeding equipment. Since there is only minimal polymer

feeding equipment associated with the application of the water soluble dispersion polymers, treatment of effluents from agricultural waste processing, the dairy and brewing industries, and metalworking and related operations, to list a few examples, has been made accessible. For instance, treatment of the large amount of wastewater generated on livestock farms can now be accomplished in an economically attractive manner because of the unique dissolution properties associated with the water-based liquid dispersion polymers.

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POLY(LACTIC ACID) PROPERTIES AND PROSPECTS OF AN ENVIRONMENTALLY BENIGN PLASTIC: MELT RHEOLOGY

John R. Dorgan

Department of Chemical Engineering, Colorado School of Mines, Golden, CO 80401

INTRODUCTION

Plastic packaging materials are everywhere in our daily lives. From food wrappings and containers to detergent and soft drink bottles to foam packaging for shipping delicate goods, many products are surrounded by or contained in polymeric materials. Currently, most polymeric packaging materials are based on non-renewable fossil resources. Incineration of these materials makes a net contribution to atmospheric CO₂ and plastics currently account for in excess of 20% of the nation's landfills. In addition, many widely used materials, notably polystyrene and poly(vinyl chloride), are made from noxious or toxic monomers. Clearly, there exists a need for the development of "green" packaging materials that would be based on renewable resources, would not involve the use of toxic or noxious components in their manufacture, and could allow composting to naturally occurring degradation products or could be recycled easily. However, for acceptance in the marketplace these materials must be competitive on a cost-performance basis.

Such a biodegradable family of polymers made from monomers obtained from *renewable* sources can be produced and will soon be competitive in the marketplace. This material is poly(lactic acid) (PLA) the polymer of lactic acid which can be made by the fermentation of corn. An independent evaluation of the process economics performed at the Argonne National Laboratory has found that advances in fermentation and separation technologies allow the manufacture of lactic acid for twenty-five cents per pound and that poly(lactic acid) (PLA) is an economically feasible material. Further evidence of the technological and commercial significance of PLA is presented below.

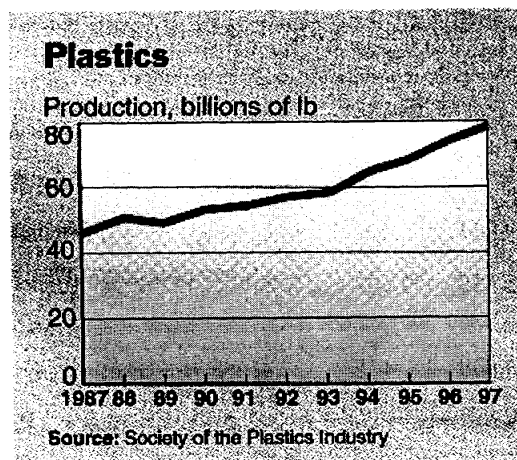
The advantages of PLAs are numerous and include: 1) production of the lactide monomer by fermentation of a renewable agricultural source (corn), 2) fixation of significant quantities of carbon dioxide, the leading greenhouse gas, 3) significant energy savings, 4) the ability to recycle back to lactic acid (a non-toxic, naturally occurring metabolite) by hydrolysis or alcoholysis, 5) the capability of producing hybrid paper-plastic consumer packaging that is compostable, 6) reduction of landfill volumes, 7) improvement of America's farm economy, and 8) the all important ability to tailor physical properties through material modifications.

SUSTAINABILITY IN THE PLASTICS INDUSTRY

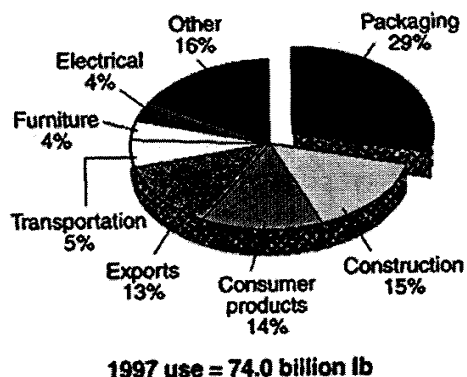
The benefits of plastic materials to humanity are considerable. Plastic packaging assists in maintaining purity and reducing spoilage in the food supply. The use of protective plastics during shipping reduces damage during transit and thus saves materials and energy needed to repair or replace goods. Because plastics are processed at low temperatures and are easily shaped, durable manufactured goods require less energy input. Another example of the beneficial use of plastic materials is in the health field where sterility is important and one-time use products are required (blood bags, heart-lung oxygenators, etc.). Plastic materials improve the quality of life for each of us and technologies are eventually needed that will assure a continued supply of inexpensive, readily processed, plastic materials for a host of important disposable applications.

Significant issues of sustainability exist in the plastics industry based on its current structure. All major commodity plastics are presently based on petroleum resources. Proven petroleum reserves are about one trillion barrels. Depending on the assumptions made regarding the rate of industrialization in the third world and growth in the developed world, this supply may last for as long as a few hundred years. As the supply of petroleum depletes, its cost will again rise and the present low cost of fossil based plastics will not be possible. Figure 1 shows the growth in thermoplastic production in the United States for the past ten years; plastic use has approximately doubled every ten years for the past three decades.

Figure 1. Growth of thermoplastics production in the United States



Packaging is top thermoplastic use



Present US consumption of 80 billion pounds per year can be expected to increase. It is evident that a doubling of fossil based plastics consumption each decade in light of declining petroleum reserves is not sustainable.

Figure 2 demonstrates that disposable packaging represents nearly one-third of all plastics use. This means that present consumption patterns result in the use of in excess of 25 billion pounds of non-renewable plastics each year for one time, throw away, applications. Recycling can help, but acceptance is poor (and not allowed in food applications) because of property degradation.

Figure 2. Packaging accounts for 29% of thermoplastics use.

Another issue of sustainability faces the plastics industry. Namely, emission of carbon dioxide from fossil fuel burning is considered as a potential threat in the form of global warming. As is the case for all industrial processes, the energy input required normally results in the production of carbon dioxide. Figure 3 compares the projected concentration of CO₂ to that needed in order to stabilize the atmospheric concentration. While not unique to the plastics industry, if the threat of global warming is to be taken seriously steps must be taken to reduce CO₂ emissions. Burning plastics to recover the energy content contribute to carbon dioxide emissions. The use of plastics packaging should be scrutinized for potential CO₂ savings.

Finally, it should be noted that plastic materials represent a serious solid waste disposal problem. EPA estimates that 20% by volume of municipal landfills are occupied by plastics. This is an even greater concern in Europe and Japan where landfill space is not as available as in the United States.

Several concerns arise regarding the sustainability of the plastics packaging industry. While the situation is not yet critical, it is prudent to work now towards the creation of more sustainable technologies.

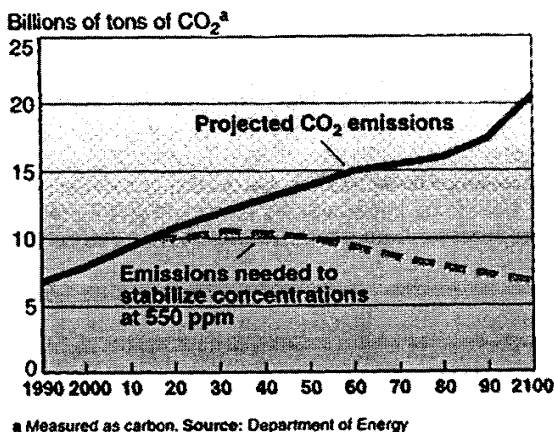
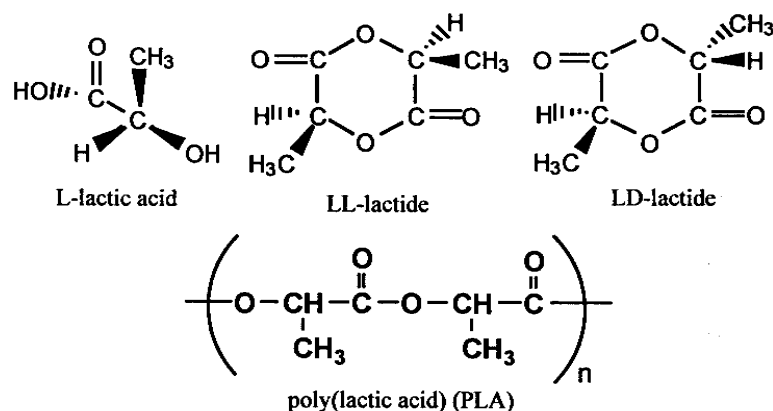


Figure 3. Projected and needed carbon dioxide emissions.

PROPERTIES OF PLA

In developing a sustainable technological basis for the plastics industry, several factors must be considered. First, plastics are carbon based and the unique bonding and atomistic features of carbon can probably not be replaced by any other element. It is important that the proposed route to sustainability de-emphasize petroleum and strive for environmental benefits and energy savings. The Department of Energy estimates that global energy capture by photosynthesis of plant matter (biomass) is about 7×10^{17} kcal/yr. but only 7% is utilized by mankind. Sensational changes in applied agronomy brought about by dramatic advances in molecular biology hold the promise of using plants as a way of producing needed carbon-based chemical feedstocks. Combined with new technologies in fermentation and chemical separations, these advances will lead to the significant utilization of bio-based materials in the place of traditional petrochemicals. Poly(lactic acid) (PLA) made from a corn-based monomer is poised to become the first large scale commercial success of this pioneering approach.



PLA is produced by milling and fermenting corn to produce lactic acid. Conventional chemical processing is used to convert the lactic acid to a lactide monomer. This monomer may be polymerized to produce a high molecular weight polymer having good transparency and strength. Figure 4 shows the chemical structures of L-lactic acid, the available monomers, and the polymer chain structure.

Figure 4. Structure of PLA and its precursors.

The benefits of PLA are manyfold. It is not based on fossil fuels and its renewable nature means that it is sustainable as long as agricultural production of corn is sustained. The amount of greenhouse gas emitted is reduced because of the fact that the carbon in the polymer comes from fixation of atmospheric CO₂. Less energy is used to manufacture PLA than competing products like polystyrene. PLA is degradable so it can be recycled back to its monomer by simple hydrolysis. Use in hybrid paper-plastic packaging provides a material that can be composted so solid waste handling and municipal landfill volumes can be reduced. The results of a life cycle analysis comparing the production of 300 million pounds of polystyrene to the same amount of PLA demonstrates that the energy and CO₂ emission savings of PLA are profound.

As mentioned above, acceptance of PLA will be based on performance. Fortunately, PLA does possess good physical properties that are in fact, very similar to polystyrene. A comparison of physical properties of PLA to other commodity plastic is given in Figure 5.

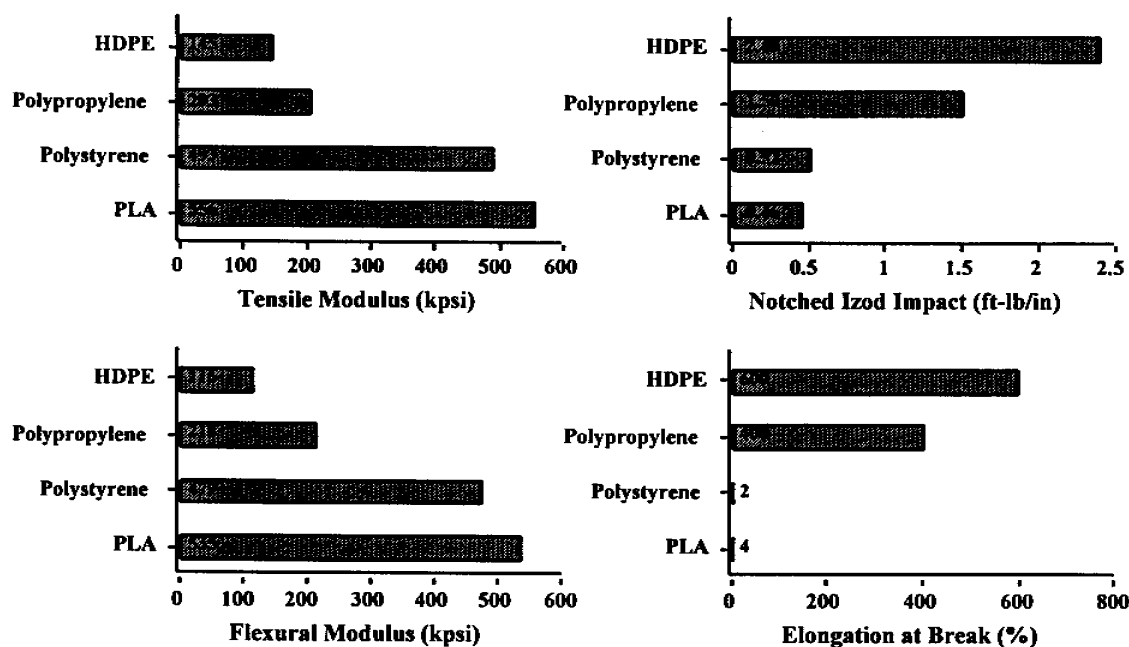
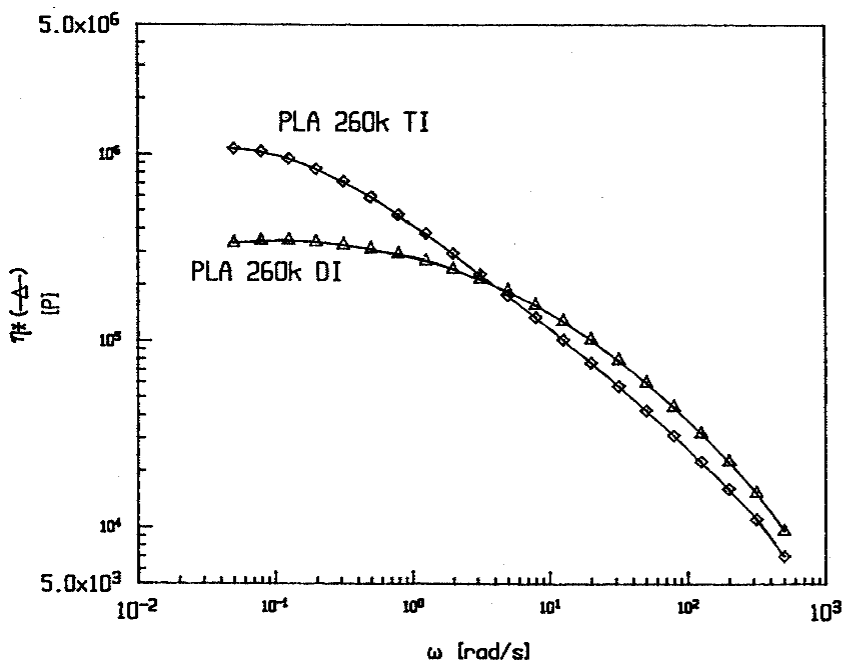


Figure 5. Physical properties of PLA compared to other plastics.

Physical properties, while important in end-use applications, do not tell the entire story. For economic competitiveness, the plastic material must also have good processing characteristics that allow rapid manufacturing of goods. Plastics processing takes place in the melt state so understanding the flow properties (the rheology) of PLA is important. Changing the molecular architecture of the polymer by introducing branching is a well-known route to altering flow properties. In this work, the first study of such molecular variations on the rheology are reported. In particular, the star architecture for PLAs was investigated. Unlike a linear polymer chain, star polymers consist of a core site with a variable number of protruding arms. Six and four arm star molecules of differing molecular weights were studied and compared to the linear material. Figure 6 shows a direct comparison of the flow properties of two PLA materials having the same nominal molecular weight (260 k) — the star material has a higher viscosity at low shear rates but a lower viscosity at high shear rates. This flow behavior is advantageous for some forming operations.

Figure 6. Viscosity versus shear rate for different molecular architectures of the same molecular weight. The sample labeled "TI" is a tetrol-initiated 4-arm star while the "DI" sample is a linear chain.



This study has shown that flow properties can be profoundly altered without sacrificing mechanical properties in PLA. This work will be continued to investigate blends of linear and branched PLAs as a means of customizing processing behavior. Such information is needed in order to improve the competitiveness of PLA and thus promote its use as a replacement for fossil based plastic packaging materials.

PROSPECTS OF PLA

The prospects of PLA are excellent. It has the same ability for property manipulation through architectural variation and copolymerization as do other highly successful plastics. In addition, it offers significant environmental benefits. As such, it is the first example of the transition of the plastics industry towards a sustainable future. This future will only be possible through additional technical effort to engineer the mechanical, flow, permeation, adhesive, and additional properties of PLA and other future plant-based plastics.

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WATER-BASED POLYESTER/ACRYLIC COATINGS VIA HYBRID MINIEMULSION POLYMERIZATION

John G. Tsavalas, Jan W. Gooch and F. Joseph Schork
School of Chemical Engineering, Georgia Institute of Technology, Atlanta, GA 30332-0100

Hybrid miniemulsion polymerization was performed with a three component acrylic system of methyl methacrylate, butyl acrylate, and acrylic acid in the presence of a Bayer• Roskydal TPLS2190 unsaturated polyester resin. Novel latexes were obtained in which the polyester resin was grafted to the acrylic polymer, forming a water-based crosslinkable coating. Both emulsions and latexes were shelf stable for over 6 months. Resin to monomer ratios were studied as high as 1:1 (wt:wt) and total emulsion solids as high as 45%. Monomer droplet and latex particle sizes were similar suggesting evidence of the preponderance of droplet nucleation. A high level of crosslinking (> 70%) during polymerization was observed in this particular hybrid system in contrast to those involving alkyd⁵ or polyurethane⁶ resins (< 5%). Films, both homogeneous and hard, were achieved with exceptional adhesion. Electron microscopy showed the hybrid particle.

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**PULP
AND PAPER**

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NEW AQUEOUS CATALYSTS FOR REPLACING CHLORINE BY OXYGEN IN WOOD-PULP BLEACHING

I. A. Weinstock, C. L. Hill, E. M. G. Barbuzzi, S. E. Reichel, R. Heintz, J. J. Cowan,
W. E. Morgan, R. S. Reiner, D. M. Sonnen, C. J. Houtman, E. L. Springer and R. H. Atalla
USDA Forest Service, Forest Products Laboratory, Madison, WI 53705
and the Department of Chemistry, Emory University, Atlanta, GA 30322

A two-step process involving selective delignification and catalytic waste mineralization could make it possible to replace chlorine by oxygen in the bleaching of chemical wood pulps. The technology uses early transition metal oxygen anion clusters (polyoxometalates or POMs) in a catalytic process that uses oxygen and operates in water. Over the past year, a top candidate catalyst for use in the new technology has been developed. The control of oxygen reactivity and of catalyst behavior in water (the most desirable yet challenging solvent for operation of transition-metal catalysts) is exemplified by fundamental studies concerning this new aluminum-based POM catalyst system.

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POLYMER-BASED AQUEOUS BIPHASIC EXTRACTION TECHNOLOGY IN THE PAPER PULPING PROCESS

Jonathan G. Huddleston, Heather D. Willauer, Scott K. Spear, Kimberly D. Smith and Robin D. Rogers
Department of Chemistry, The University of Alabama, Tuscaloosa, AL 35487

The chemical pulping of wood represents the major route to the production of paper and board. Current processes are highly efficient in terms of chemical use but are beset by numerous environmental problems. Successful reengineering of this process through the use of organic solvents to solubilize lignins from the cellulosic fraction of wood has been achieved in the "Organosolv Process". This process has not been adopted on any significant scale because of the difficulties of engineering high temperature processes involving organic solvents. Numerous polymers are known which phase separate under the influence of temperature, salt, or in the presence of other polymers and which represent efficient extractants of organic compounds. In this way the use of organic solvents could be avoided. Preliminary work in attempting to develop paper pulping as an aqueous biphasic extraction process will be presented.

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TAML™ CATALYTIC OXIDANT ACTIVATORS IN THE PULP AND PAPER INDUSTRY

Terry Collins
Department of Chemistry, Carnegie Mellon University, 4400 Fifth Avenue, Pittsburgh, PA 15213

Catalysis will play a key role in improving four major features of wood pulp bleaching in the pulp and paper industry in the twenty-first century. These features are:

- capital and operating costs
- product quality
- environmental performance
- energy efficiency

The importance of chemical selectivity as it applies to bleaching wood pulp will be reviewed and the significance of the difference in energy profiles between catalyzed and uncatalyzed bleaching reactions will be explained. Seven promises of catalyzed activation of hydrogen peroxide for pulp bleaching will then be presented. These promises will be illustrated by describing the state-of-the-art of TAML™ bleaching technology. TAML™ technology will be presented as a highly selective, totally-chlorine free method of bleaching kraft pulp. The promise of the new technology will be described to achieve high strength pulp while delivering the following advantages:

- saving energy in the bleach plant
- reducing or eliminating color in pulp mill effluent steams
- reducing or eliminating persistent pollutants in pulp mill effluent steams
- improving the efficiency of peroxide usage
- eliminating future requirements for large capital expenditures in bleaching

Design work will be described aimed at producing special purpose catalysts to approach "ideal behavior," wherein it is proposed that activated peroxide will react in a highly selective manner to oxidize lignin with little or no destruction of the cellulosic structures.

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